December 2020

UCDAVIS
VETERINARY MEDICINE
Center for Equine Health

2020 CENTER FOR EQUINE HEALTH

RESEARCH REVIEW

New Discoveries in Equine Health

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2020 CENTER FOR EQUINE HEALTH RESEARCH REVIEW

New Discoveries in Equine Health – December 2020

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Published by the Center for Equine Health

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On the cover:

Cover artwork by Carl Nelson Gorman from 1960, gifted in memory of Shirley and Norman Carroll O'Connor to the UC Davis Museum that bears his name.

The C.N. Gorman Museum, currently located on the UC Davis campus in Hart Hall, was founded in 1973 and is dedicated to the exhibition and engagement with contemporary Native American and Indigenous art. For almost half a century the C. N. Gorman Museum's reputation of artistic excellence has been proven by its company of exhibiting artists, both established and emerging, and dedication to cultural diversity.

Director's Message

Greetings!

It is with great excitement that I welcome you to the 2020 Research Review, focused on the research accomplishments of UC Davis faculty, residents, students and staff as supported by the Center for Equine Health (CEH). The Center's mission is to support equine teaching, research and service activities essential to the UC Davis School of Veterinary Medicine. Our researchers have addressed challenging questions in efforts to advance equine health and welfare. This Review is a summary of these findings, translating our research discoveries directly to horse owners and veterinarians.

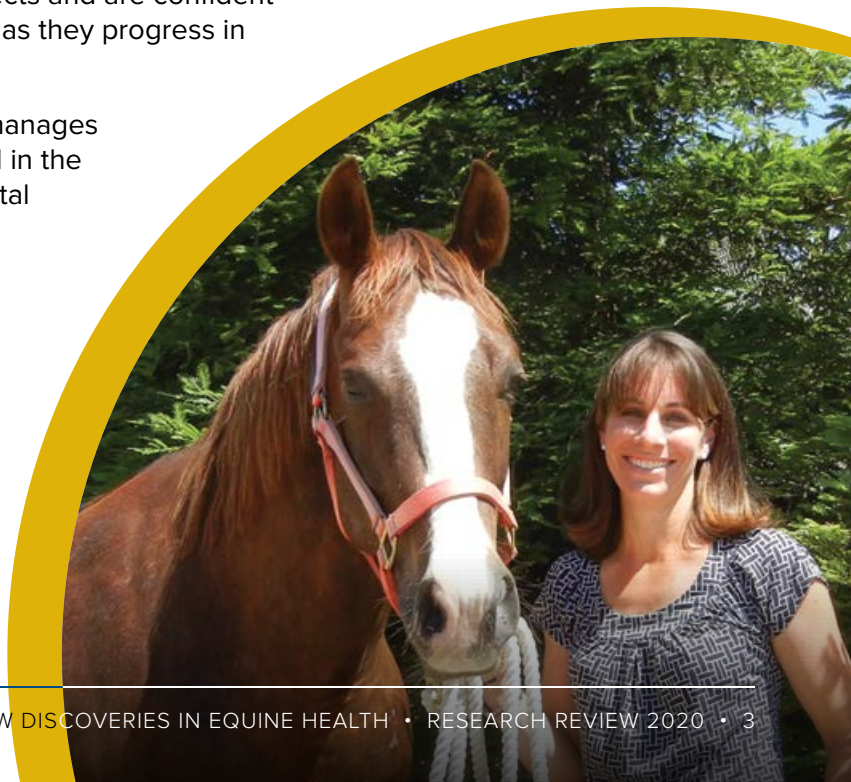
Among these findings, UC Davis researchers have made significant strides in injury prevention in racehorses. Dr. Susan Stover and colleagues at the J.D. Wheat Orthopedic Laboratory presented results on racetrack surface properties and the role of proximal sesamoid bone fractures in catastrophic breakdowns in racing Thoroughbreds. Dr. Mathieu Spriet and collaborators pioneered the ***first equine positron emission tomography (PET) scan*** and showed that the technology can successfully detect injuries at the molecular level. This diagnostic modality is currently being used in our southern California racetracks to PREVENT racehorse injuries.

This Review also highlights the outstanding research performed by UC Davis veterinary residents. Resident and graduate training are top priorities for CEH in order to train the next generation of equine clinician scientists. The CEH grant program provides residents with firsthand experiences conducting research projects, from writing grants to generating results, and analyzing and publishing their findings. We are proud to share the outcomes of resident research projects and are confident that these young professionals will make important contributions to veterinary medicine as they progress in their careers.

In addition to providing research funding through our competitive grant program, CEH manages a teaching herd of horses that are at the ***heart of our efforts***. The discoveries presented in the following pages would not be possible without our equine partners. We recognize the vital roles that these exceptional equines play and celebrate their contributions to improving health for all horses.

Our research is supported by the generosity of the many donors who continue to make the Center's research program a success. ***Thank you*** to each and every donor for your investment in CEH for the health and well-being of horses.

Carrie J. Finno, DVM, Ph.D., DACVIM
Director, Center for Equine Health



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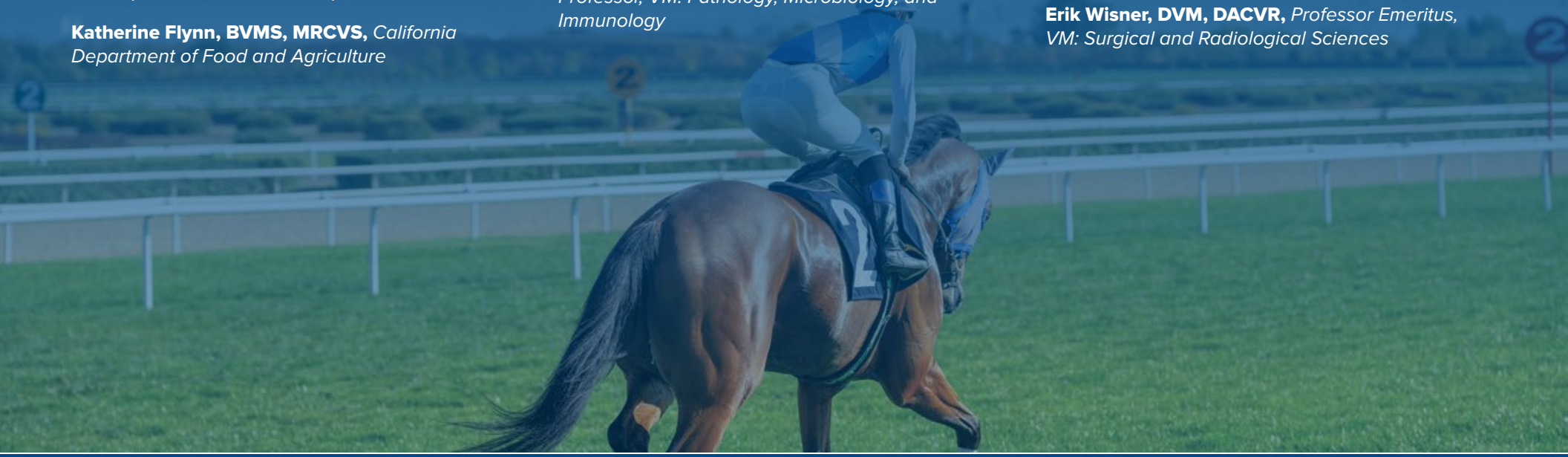
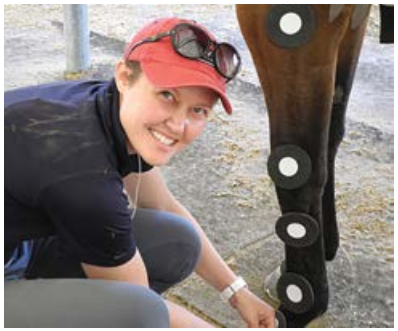


CENTER FOR EQUINE HEALTH AWARDS

2018 James M. Wilson Award – Jennifer Symons, PhD



Dr. Jennifer Symons received the 2018 James M. Wilson Award in recognition of her contributions to equine research. As a mechanical and biomedical engineer, she collaborated with Dr. Sue Stover and other researchers on the study "Modelling the effect of race surface and racehorse limb parameters on in silico fetlock motion and propensity for injury."

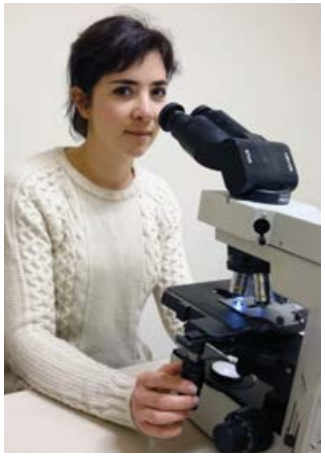
The aim of this study was to consider the effect of changing factors on fetlock motion during gallop, and to determine which factors produce the greatest changes. Dr. Symons and her colleagues tested this using a computer model of a virtual racehorse galloping on a virtual race surface. Study results indicate that the depth of the upper, softer layer of the race surface has the greatest potential to influence fetlock flexion. Increasing the depth of this layer within the model decreased the degree of simulated fetlock flexion during gallop. Practically, this parameter is related to race surface maintenance, specifically depth of harrowing. Other parameters that produced lesser changes in fetlock motion included lower layer race surface mechanics and racehorse tendon/ligament stiffness. Changes in friction between the hoof and race surface produced the smallest changes in fetlock motion.

These computer model results provide evidence to guide race surface management decisions to reduce the incidence of fetlock injuries in racehorses, particularly through the depth of harrowing race surfaces.

James M. Wilson Award

The James M. Wilson Award is given each year to an outstanding equine research publication authored by a graduate student or resident in the UC Davis School of Veterinary Medicine. The Center for Equine Health Scientific Advisory Board judges the papers based on scientific merit, quality of writing and relevance to the equine industry. Dr. Wilson was a 1945 graduate of the Ohio State University College of Veterinary Medicine. He was a well-known and respected racetrack veterinarian in California and maintained a strong interest in equine research at UC Davis.

2019 James M. Wilson Award – Regina Zavodovskaya, DVM



Dr. Regina Zavodovskaya received the 2019 James M. Wilson Award for her research publication describing signals of bone production in horses with silicate-associated osteoporosis. Dr. Zavodovskaya is a board -certified veterinary pathologist pursuing a PhD in the Integrative Pathobiology Graduate Group under the mentorship of Dr. Susan Stover.

Osteoporosis is a debilitating disease in which bones are weakened and prone to disfigurements and fractures. Horses with fragile bones have been identified in regions of California with soils rich in naturally occurring toxic silicate crystals. These horses also suffer from a lung disease called silicosis as a result of breathing dust containing the crystals. Concurrent osteoporosis and silicosis is named silicate-associated osteoporosis (SAO).

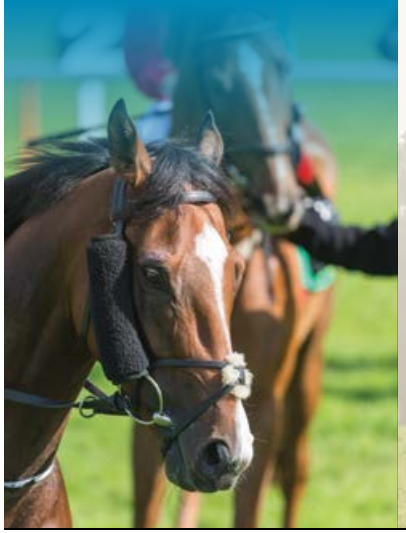
Since there are currently no markers for diagnostic testing, Dr. Zavodovskaya investigated the mechanism of bone loss by comparing tissue signal profiles in bones of horses with and without SAO. Computer programs assessed the signal patterns and constructed signal profiles differentiating SAO-affected and normal horses. The study results showed an unexpected pattern of increased bone production in SAO horses compared to the normal group. It is possible that affected horses produced more bone as compensation for the excessive bone destruction that they experienced.

These results provide signals specific to SAO that can be developed into a much-needed tool to detect affected horses early and slow the progression of SAO through appropriate treatment.

CENTER FOR EQUINE HEALTH AWARDS

Louis R. Rowan Fellowship

The Louis R. Rowan Fellowship, funded by the California Thoroughbred Foundation with financial assistance from the Oak Tree Racing Association, was established in memory of one of the California Thoroughbred Foundation's founders. In addition to being a noted racehorse owner and breeder, Rowen was active in many areas that benefited people and horses in the Thoroughbred world.



2018 Rowan Fellowship – Monica Pechanec

Monica Pechanec was selected to receive the 2018 Louis R. Rowan Fellowship. She is a Ph.D. student in the Animal Biology Graduate Group conducting research on equine tendon regeneration under the guidance of Dr. Michael Mienaltowski. She is combining the fields of anatomy, engineering, biochemistry, and molecular biology as she researches tendon formation in horses. The goal is to develop insights and strategies for equine tendon repair.

She did her undergraduate work at Brown University and earned her M.S. in animal biology at UC Davis. In addition to her research work, she has served as a teaching assistant at UC Davis.

Outside her university pursuits, she is a horse enthusiast and athlete. Since relocating to Davis, she acquired a racing Arabian, which she trains for long trail rides and possible endurance riding. She was a childhood gymnast and competed in pole vault at Brown.

2019 Rowan Fellowship – Sarah Shaffer

Sarah Shaffer, a Ph.D. candidate in mechanical engineering, was awarded the 2019 Louis R. Rowan Fellowship. Shaffer conducts research on racehorse fractures at the J.D. Wheat Veterinary Orthopedics Research Laboratory under the direction of Dr. Susan Stover.

Fractures of the proximal sesamoid bones, which are located in a horse's ankles, are among the most common fatal bone fractures in Thoroughbred racehorses. The goal of Shaffer's work is to create a mathematical model of the proximal sesamoid bones that will predict fracture risk based on the training program. Ideally, results could provide recommendations to owners, trainers, and veterinarians as to which training programs put horses at a higher or lower fracture risk.

A lifelong equestrian, Shaffer holds a National Reining Horse Association Youth World Title and multiple top ten placings at the American Quarter Horse Congress in reining with her Quarter Horse, Roostamatic (a.k.a. Beau). After completing her Ph.D., Shaffer intends to work in the equine biomechanics field as a professor at a research institution.

RESOURCE FUNDS

Center for Equine Health Directorship Support Fund – This fund was established by the estate of Joyce E. Williams in 2015 to provide support funds to the CEH director.

Director's Endowment – The Director's Endowment provides general funding for Center for Equine Health research, educational or welfare activities most critical to the needs of the horse in any given year. This endowment also provides the foundation for all Center endeavors.

Gregory L. Ferraro Endowed Directorship – This endowment was established in 2015 in honor of Dr. Gregory L. Ferraro, director emeritus of the Center for Equine Health, for his lifelong dedication to advancing the health and welfare of horses. The fund provides support to the director to develop the vision and plan for the enduring success of the Center.

Polly and Bill Swinerton Director's Endowment – This fund supports the activities of the Center for Equine Health Director to advance the facility's teaching, research and service missions.

William and Inez Mable Family Foundation Endowment – This endowed fund was established to support the Center for Equine Health in its operational, educational and research efforts. Endowment earnings are distributed at the direction of the Center Director for advancing the health, well-being, performance, and veterinary care of horses through research and/or education.

INNOVATION FUNDS

Animal Rescue and Disaster Medicine Endowment – The Animal Rescue and Disaster Medicine Endowment supports the development of improved techniques for the rescue of large animals during natural disasters. The fund also provides for research into various medical conditions of the animals and the development of improved treatment regimens.

Bernard and Gloria Sallck Equine Viral Disease Laboratory Endowment – This endowment supports a program dedicated to international scientific investigations of emerging equine viral diseases. Its goal is to identify and control viral diseases of the horse that can affect the international movement, commerce and health of competitive equine athletes.

Dan Evans Memorial Endowment – The Dan Evans Memorial Endowment provides funding for UC Davis Veterinary Medical Teaching Hospital resident house officers to conduct research in any area of equine medicine and surgery that is relevant to the development of their specialty board certification.

Enduring Legacy Endowment – The Enduring Legacy Endowment was established to provide for the administration of experimental or high-risk therapies to severely ill or injured horses with unique veterinary conditions for which there is a high degree of learning value associated with their condition. The fund also supports the clinical trials program within the School of Veterinary Medicine.

Equine Athletic Performance Laboratory Endowment – The Equine Athletic Performance Laboratory Endowment provides for the development of analytical methods for accurately evaluating the athletic conditioning and performance capability of individual horses. Once these analytical techniques are fully developed, the goal of the program is to provide an objective evaluation of the ability of drug agents and training methods to enhance performance and decrease the risk of injury in competitive horses.

James M. Wilson Endowment – Established in 1995 to honor Dr. James M. Wilson, the fund supports an annual award recognizing an outstanding resident or graduate student researcher.

J.D. Wheat Veterinary Orthopedic Research Laboratory Endowment – The J.D. Wheat Veterinary Orthopedic Research Laboratory investigates the underlying causes of bone fractures, their prevention, and new methods of fracture repair. This Laboratory was originally established by the Southern California Equine Foundation, Inc., with funds provided by the Dolly Green Research Foundation.

John P. Hughes Memorial Endowment – Named after the founding director of the Center for Equine Health, the John P. Hughes Memorial Endowment provides funding for UC Davis Veterinary Medical Teaching Hospital resident house officers to conduct clinical research in any area of equine medicine or surgery.

INNOVATION FUNDS continued

Juliette Weston Suhr Fellowship Fund – The Juliette Weston Suhr Fellowship is awarded to postgraduate veterinary students who are interested in conducting research in the areas of exercise-related cardiopulmonary and metabolic disorders.

Lorna Talbot Equine Biomedical Fund – Established by Lorna Talbot in 2003, the fund promotes the development of new and re-emerging research programs in basic equine sciences.

Lorna Talbot Equine Clinical Program – Established by Lorna Talbot in 2003, the fund promotes the development of new medical programs of clinical relevance with the Veterinary Medical Teaching Hospital.

Lucy G. Whittier Endowment for Equine Perinatal and Infectious Disease – The Lucy G. Whittier Endowment is dedicated to improving the health and medical treatment of newborn foals and their dams and to conduct research on infectious diseases associated with foals.

Patricia J Hobbs Endowed Research – Established by the estate of Patricia J Hobbs in 2009 to support research in the field of equine laminitis.

Patricia Yeretzian Endowment Fund – This fund was established by longtime Silver Stirrup Society members, Patricia and Paul Yeretzian. The fund supports equine research projects relating to reproduction and infertility disorders.

Peray Memorial Endowment – The Peray Memorial Endowment is an important resource for resident house officers of the UC Davis Veterinary Medical Teaching Hospital to conduct equine respiratory disease and colic research.

Performance Horse Endowment – Medical problems of the mature show and event horse are the focus of the Performance Horse Endowment. This endowment also funds long-term, in-depth studies of problems that preclude horses from performing athletically as they age. Areas of study include colic, nutrition, cardiopulmonary health, degenerative orthopedic processes and infectious disease.

Sundance Ranch Endowment – This fund was established by the late Carol Green to provide funding support for research in biological and translational research in the pursuit of effective treatments and cures for systemic diseases of the horse. Ms. Green had particular interest in medical conditions related to the development of laminitis.

Simulcast Racing Contributions

In 1987, the Satellite Wagering Act (Senate Bill 14) designated one-tenth of one percent of California's simulcast racing handle to be used for equine research. In 1994, Senate Bill 518 was passed, designating the redistribution of the simulcast racing percentage. These funds support both the Center for Equine Health and the Kenneth L. Maddy Equine Analytical Chemistry Laboratory. This important laboratory has three components: (1) a full-service, routine drug testing program, (2) a forensic toxicology program, and (3) a pharmacology research and methods development program. The latter includes the development of new tests and documentation of drug testing effects on racehorse performance. In 2001, the Account Wagering Bill (Assembly Bill 471) was passed, directing simulcast contributions made through televised wagering to UC Davis equine research and drug testing programs.

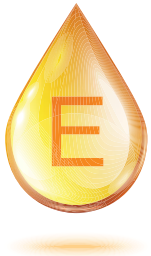
COMPLETED RESEARCH STUDIES

DRUG THERAPIES

Investigation of vitamin E metabolism in horses and the effect of supplementation (Grant #16-14)

Investigators: Birgit Puschner, DVM, PhD, DABVT, Carrie J. Finno, DVM, PhD, DACVIM, Xiaopeng Chen, PhD

Vitamin E (vitE) is one of the most important fat-soluble vitamins and horses obtain most of their vitE from fresh pasture. Understanding the metabolic pathways of vitE is essential to define optimal dietary vitE intake in horses. Additionally, the cause for equine neuroaxonal dystrophy (eNAD) remains unknown but may be due to altered vitE metabolism. We were able to develop reliable assays to detect concentrations of vitE isoforms (α-tocopherol, γ-tocopherol, α-tocotrienol, γ-tocotrienol) and metabolites (α –CMBHC, α –CEHC and γ-CEHC) in serum and urine from healthy horses with adequate access to fresh pasture. Once we established reference ranges, we assayed them in a group of healthy horses and eNAD-affected horses before and after administration of 5000 IU of α-tocopherol orally. We found that horses with eNAD metabolized α-tocopherol more rapidly than unaffected horses. These results support the current hypothesis that the etiology of eNAD involves a genetic defect leading to abnormal metabolism of vitamin E.



How does this research benefit horses? We have developed an assay to measure concentrations of vitE isoforms and metabolites in equine serum and urine samples. Preliminary results from this study indicate that this could potentially serve as an antemortem diagnostic test for eNAD; however, we will first validate these findings in a larger study population.

This research is in press with the *Journal of Veterinary Diagnostic Investigation.*

GENETICS

Investigating a genetic risk factor for eye and skin cancer in horses (Grant #16-06)

Investigators: Rebecca R. Bellone PhD, Mary Lassaline DVM, PhD, DACVO, Christopher M Reilly MAS, DVM, DACVP, Tammy Miller Michau DVM, MS, MSpVM, DACVO, Jiayin Liu, Savanna Vig DVM, Moriel Singer-Berk BS

Squamous cell carcinoma (SCC) is the most common cancer of eyes and the second most common tumor of the horse overall. A change in the DNA that also leads to a change in the gene product (missense mutation) in *damage-specific DNA-binding protein 2* (DDB2, c.1012 C>T, p.Thr338Met) was previously found to be strongly associated with SCC in Haflingers. This study determined that this same mutation explains 76% of all ocular cases of the Haflinger and Belgian breeds. This variant also explained ocular SCC in the only reported case from the Rocky Mountain Horse breed. This variant was not associated with ocular SCC in the Arabian, Appaloosa, or Percheron breeds, or with oral or urogenital SCC of any breed investigated (P>0.05). Finally, functional analysis using recombinant DDB2 protein further supports that this variant (p.Thr338Met) is a causal genetic risk factor, as the protein with methionine at amino acid 338 is unable to bind to UV damaged DNA.

How does this research benefit horses? This study confirmed that the *DDB2* mutation is a risk factor specifically for ocular SCC in multiple breeds of horses (Haflinger, Belgian, and Rocky Mountain Horses, but not Appaloosa and Arabian). This further supports the use of this mutation as a breed specific DNA test to inform mating decisions and management practices, thus lowering the incidence of the disease, enabling earlier detection and better prognosis.

This research was reported in *Animal Genetics* 2018 Oct; 49(5):457-460, *Veterinary Ophthalmology* 2019 Mar; 22(2):201-205, the *International Journal of Genomics* 2019 Sept; 3610965, and the *Equine Veterinary Journal* 2020 Jan; 52(1):34-40.

To obtain additional information on a research study, please reference the grant number in your request.

Investigating genetic risk loci for ocular squamous cell carcinoma in horses (Grant #17-12)

Investigators: Rebecca R. Bellone, PhD, Mary E. Lassaline, DVM, PhD, Kelly E. Kniceklbein, VMD

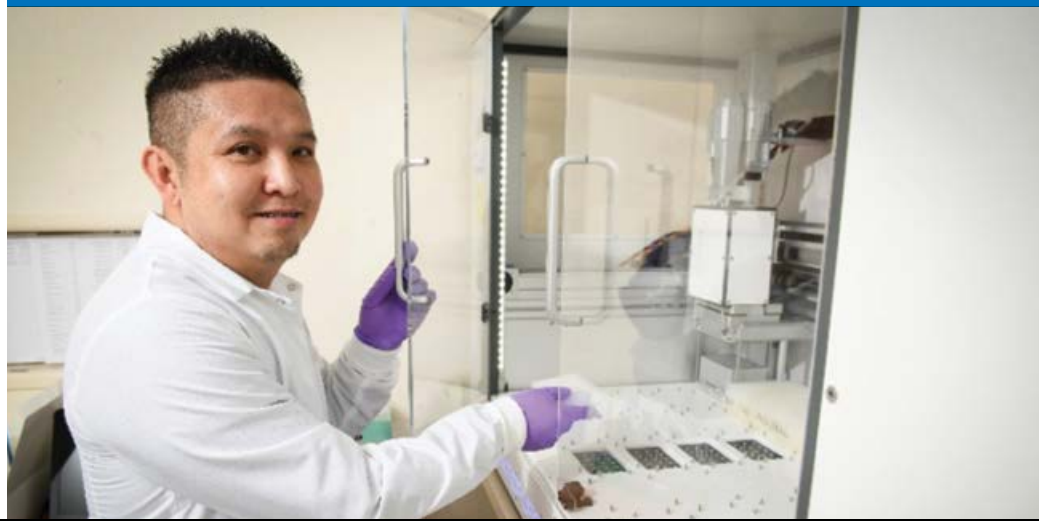
Drs. Mary Lassaline and Kelly Knickelbein examining a Haflinger horse to enroll in the ocular SCC study.

Squamous cell carcinoma (SCC) is the most common cancer of the equine eye and the second most common tumor of the horse overall. SCC frequently originates on the third eyelid or the limbus (where the clear cornea meets the white sclera) and can grow quickly to invade the eye and adjacent structures, leading to vision loss and destruction of the eye. A mutation in a DNA ultraviolet radiation repair enzyme (*DDB2*) was previously identified as associated with increased risk for ocular SCC development in the Haflinger and Belgian breeds, though not all affected horses could be explained by this mutation. The objective of this study was to identify additional DNA mutations for association with ocular SCC in Haflinger and Belgian horses. Investigating the previously associated locus did not identify another variant more strongly associated with ocular SCC in Haflingers or Belgians than the one identified previously in *DDB2*. When taking the *DDB2* risk variant genotype into account, statistical analysis supported a variant located on horse chromosome 6 as a second risk factor for ocular SCC in both Belgian and Haflinger horses (P=2.75 X10-4). This variant is a missense variant in a gene involved in the same UV damage DNA repair process as *DDB2*.

How does this research benefit horses? This work has advanced our understanding of the genetic risk for ocular squamous cell carcinoma in horses. These data further support the use of genetic testing for the *DDB2* variant in Haflinger and Belgian breeds. Testing for this variant can inform management and breeding decisions. This study also identified an additional genetic risk factor that could be utilized in DNA testing to develop the most robust ocular SCC risk assessment in these two breeds. Functional studies are needed to confirm the role of this variant in cancer.

This research was reported in the *International Journal of Genomics* 2019 Sept; 3610965.

THE UC DAVIS VETERINARY GENETICS LABORATORY is now offering a genetic test for Ocular Squamous Cell Carcinoma (SCC) in Haflinger, Belgian and Rocky Mountain Horses. Testing for this risk variant can help owners and breeders identify horses at higher risk and can assist in mating pair selection. Homozygous horses (R/R) are advised to have routine eye exams performed by a veterinary ophthalmologist for early detection and better prognosis, and to wear a UV protecting fly mask when out during the daylight hours. Breeding homozygotes (R/R) and heterozygous (R/N) should be avoided to reduce the chances of producing horses that have a high risk of developing this cancer.



Identification of the mutations resulting in blood group A factors to prevent or plan for neonatal isoerythrolysis (Grant #16-12)

Investigators: Robert A. Grahn, PhD, M. Cecilia Torres Penedo, PhD

Blood group factor incompatibilities between mares and foals require immediate postpartum care. If untreated, they can result in the death of the foal as antibodies in the mare's colostrum attach to the foal's red blood cells, causing them to be destroyed. This research evaluated one genomic location with a blood factor (Aa) known to result in neonatal isoerythrolysis (NI). Twenty-one horses were genotyped across the genomic region associated with blood factor A, including obligate blood group factor Aa homozygous horses. These data did not identify a chromosomal segment segregating with the NI associated blood group factor Aa. These data prompted the complete genome sequencing of eight horses with known blood types. Five were type Aa, three were not Aa. The horse reference genome was included in the analysis and is also not type Aa. Again, no causal variants were identified in the region containing the A blood group. Evaluation of the remainder of the genome did not reveal any significant genetic variants in genes associated with cell surface proteins and carbohydrates. It is likely that the surface protein modification resulting in the Aa blood factor is caused by a gene at another location within the genome, but additional samples are required to refine the number of possible causal variants.

How does this research benefit horses? The goal of this project was to identify causative mutations for the horse blood group A factors. As the majority of NI cases involve blood factor Aa incompatibilities, knowledge of stallion and mare genotypes at the A system has the potential to avoid mating, or minimally, anticipate and prepare for postpartum care required for NI foal survival. Identification of DNA variants associated with blood group factors involved in NI would also eliminate the current dependency on antibody reagents for serological typing. Production of reagents has virtually ceased worldwide, and future typing is limited to available stocks. The genome sequence data generated in this study has formed the basis of continued research efforts to identify the causal mutation as additional horses with known blood types are sequenced.

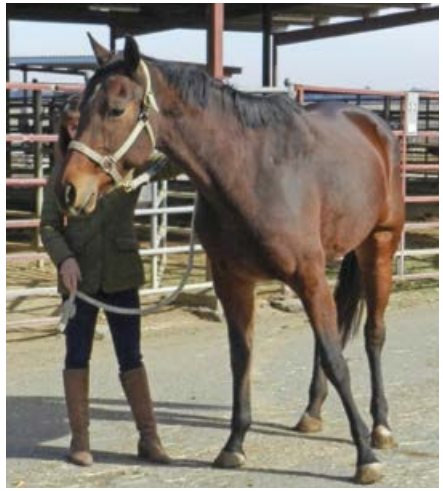
Finding the genetic mutation for equine neuroaxonal dystrophy (Grant #17-02)

Investigators: Carrie J. Finno, DVM, PhD, DACVIM

Equine neuroaxonal dystrophy (eNAD) is a devastating neurological condition that develops during the first year of life in genetically predisposed foals maintained on a vitamin E deficient diet. Affected horses suffer from incoordination, preventing their use as riding animals. Our previous study identified possible genetic regions of interest associated with eNAD. We then used whole-genome sequencing to find possible underlying mutations in these regions in a small number of eNAD-affected and unaffected horses. This study sought to validate these potential mutations in additional horses, particularly in the candidate gene, *CYP4F2*, which encodes the protein responsible for metabolizing vitamin E. Forty variants identified as possible candidates for eNAD were excluded. While no structural variants or segregating SNPs associated with eNAD/EDM were identified in *CYP4F2*, the gene demonstrated significantly increased expression (~1.63x, P=0.02) in liver from eNAD-affected horses.

How does this research benefit horses? We have identified increased expression of *CYP4F2*, which encodes the major protein responsible for vitamin E metabolism in the liver, in eNAD-affected horses. In 2016, with CEH support, we had identified altered metabolism of α-tocopherol, the major isoform of vitamin E, in eNAD-affected horses. Therefore, increased metabolism of α-tocopherol, with upregulation of *CYP4F2*, is strongly associated with the eNAD phenotype. This supports the current recommendations for high dose vitamin E supplementation in genetically susceptible herds and further refines the search for an underlying genetic variant.

This research was reported in *Genes* (Basel). 2020 Jan 10;11(1):82



An eNAD affected horse exhibiting abnormal proprioception.

Whole-genome sequencing to identify inherited genetic alterations associated with melanoma in graying Connemara ponies (Grant #17-08)

Investigators: Alain P. Theon, DVM, MS, PhD, Carrie J. Finno, DVM, PhD, DACVIM

Melanoma in graying horses is a hereditary disease with a complex mode of inheritance. Other than the graying mutation itself, no additional genetic associations have been identified for the risk of melanoma in graying horses. This study was designed to identify new actionable genetic targets for prevention and development of targeted treatment by comparing the whole genomes of graying Connemara Ponies with melanoma and without melanoma. When comparing the early onset cases to either the unaffected cohort alone or the unaffected cohort plus the late onset cohort, a large associated region on chromosome 1 was identified. This region contained a tumor-suppressing gene that is abnormal in affected ponies when compared to unaffected ponies. This is a significant finding because the presence of abnormal tumor suppressor genes is commonly found in all cancer cells. Because mutations of tumor-suppressor encoding-genes are common genetic abnormalities associated with inherited cancers, the presence of the abnormal gene we identified is consistent with our hypothesis that early severe onset of melanoma in graying Connemara ponies is associated with a key genetic alteration.

How does this research benefit horses? Horses with greying hair coat have an 80% risk of developing melanoma. Graying ponies may develop melanoma late in life that is usually associated with a slow evolution and less likely to be life threatening. However, greying ponies that develop melanoma early in life are at high risk of dying of disease due to faster evolution and earlier metastatic spread. This represents a significant commercial loss for the industry. A susceptibility test for equine melanoma in graying horses would enable veterinarians to identify graying horses at high risk of early onset disease and monitor these individuals closely for tumor development. This genetic region will be further validated, with the intent of providing a genetic test in the future for increased melanoma susceptibility in gray horses and ponies.

Finding the genetic mutation for juvenile idiopathic epilepsy in Arabian horses (Grant #17-09)

Investigators: Monica Aleman, MVZ, PhD, DACVIM and Carrie J. Finno, DVM, PhD, DACVIM

Juvenile idiopathic epilepsy (JIE) is a disorder of Egyptian Arabian foals that causes seizures and has potential life-threatening complications, including head injury and aspiration pneumonia. The disorder is inherited, and our previous study allowed us to identify a genetic region of interest associated with JIE. We then used whole-genome sequencing to find possible underlying mutations in this region in two JIE-affected horses. This study sought to validate these potential mutations in a large number of horses.

Of the 27 variants genotyped across the ~5 Mb region on chromosome 1 (chr1), only two were associated with the JIE phenotype in the full subset of horses. These variants, in addition to the original GWAS variants, further narrowed the associated region to a 250 kb region on chr1. This associated region only included one annotated gene in the horse, *mannose-binding lectin 2* (*MBL2*), a gene solely expressed in the liver and involved in the innate immune defense. However, none of the variants were located within this gene. There was no differential expression of hepatic *MBL2* in n=1 JIE-affected Arabian vs. n=5 unaffected horses. *MBL2*

is not expressed in brain tissue from any species. Additionally, no associated variants were identified in the two flanking candidate genes, *PCDH15* and *ZWINT*.

How does this research benefit horses? We have confirmed and further narrowed the region of association for JIE onto chr1 and excluded the only protein-coding gene within this region. Next steps include using data generated by the functional annotation of the equine genome project to search for possible mutations that alter gene regulation. The ultimate goal is to develop a genetic test for JIE that breeders and veterinarians can use to prevent future cases.

This research was reported in the *Journal of Veterinary Internal Medicine* 2018 Jan-Feb; 32(1):465-468.

Investigating the genetic cause of persistent hypocalcemia in Thoroughbred horses (Grant #17-14)

Investigators: K. Gary Magdesian, DVM, DACVIM, ACVECC, ACVCP, Carrie J. Finno, DVM, PhD, DACVIM

A syndrome of idiopathic hypocalcemia has been described in Thoroughbreds foals since 1997 and appears to be inherited. All identified cases have been fatal, and a genetic test is necessary to prevent future cases. This study looked for genetic mutations in five genes that have been associated with inherited hypocalcemia in humans: calcium-sensing receptor (*CASR*), G protein subunit alpha 11 (*GNA11*), parathyroid hormone (*PTH*), glial cells missing transcription factor 2 (*GCM2*) and transient receptor potential channel melastatin 6 (*TRPM6*). Pedigree analysis revealed an autosomal mode of inheritance for idiopathic hypocalcemia in Thoroughbred foals. After evaluating whole-genome data and performing subsequent analysis, no variants were associated with the phenotype in the horses tested. Therefore, these five candidate genes are unlikely to be implicated in functional hypoparathyroidism of Thoroughbred foals.

How does this research benefit horses? Idiopathic hypocalcemia in Thoroughbred foals has a 100% mortality rate. Discovery of a genetic mutation associated with this disease would allow for testing of breeding horses for the mutation, with subsequent avoidance of mating of carriers to prevent any future affected foals. As we have excluded the most likely candidate genes, the next step is to investigate the entire genome to attempt to identify a likely causal mutation.

IMMUNOLOGY

Vaccination of horses against rabies: Do we really need to revaccinate every year? (Grant #16-08)

Investigators: W. David Wilson, DVMS, MS, Johanna L. Watson, DVM, PhD, Judy E. Edman, BS

Rabies is an AAEP-designated core disease against which all horses in North America should be vaccinated. The recommended revaccination interval for horses is 1 year, whereas the same rabies vaccines are typically administered at 3-year intervals in dogs and cats. The aim of this study was to determine how long "protective" titers persist after booster vaccination of previously primed horses to determine whether an extended revaccination interval is feasible.

In phase 1, 48 horses with an undocumented vaccination history were vaccinated against rabies. Blood samples were collected prior to vaccination, 3 to 7 weeks later, and at 6-month intervals for 2 to 3 years. Serum rabies virus–neutralizing antibody (RVNA) values were measured using the RFFI test. A RVNA value of ≥ 0.5 U/mL was used to define a predicted protective immune response. A protective RVNA value (≥ 0.5 U/mL) was maintained for 2 to 3 years in all horses inferred to have been previously vaccinated. In phase 2, sampling continued on 11 of the horses for as long as they remained available. All 11 maintained "protective" antibody levels at their last sampling time, 9.5 to 13 years after receiving their last rabies booster.

The prolonged persistence of protective rabies antibody titers after revaccination of previously primed horses strongly suggests that an extended revaccination interval of 3 years or more is justified, particularly for horses that react adversely to rabies vaccines. In these horses, strategic revaccination based on measured serum RVNA titers is rational.

How does this research benefit horses? Designation by the AAEP of rabies as a core vaccine for annual administration to horses has resulted in a substantial increase in the number of horses that receive this vaccine. The unintended negative impacts have been an increased expense to horse owners and an increase in the number of horses that experience adverse reactions to rabies vaccines. By showing that continual protection can be accomplished by administering rabies "boosters" at an interval of more than one year, this study identifies options for mitigating these unintended impacts, providing clear benefits to horse owners and to horses, particularly those that react adversely to rabies vaccines.

This research was reported in the *Journal of the American Veterinary Medical Association* 2016 Aug; 249(4): 411-8.

Serological response to equine influenza virus following boost vaccination of adult horses using different commercially available killed vaccines

(Grant #17-05)

Investigators: Nicola Pusterla, DVM, PhD, DACVIM, DAVDC-Equine, W. David Wilson, BVMS, MS, DACVIM, Bruno Karam, DVM

The best achieved protection against equine influenza virus (EIV) is based on AAEP vaccine recommendations that horses ages 4-6 months should be initially prime vaccinated with a series of 3 doses given 4-6 weeks apart, followed by boost vaccination every 6 to 12 months. Most veterinarians and owners use EIV vaccines interchangeably, despite differences in EIV strains, viral mass and adjuvant in commercially available vaccines. Our hypothesis was that a switch in EIV vaccine manufacturer requires a 2-dose vaccine series to trigger an antibody response to EIV that is similar, if not superior, to what a boost vaccination with the vaccine from the original manufacturer would induce.

We enrolled 65 healthy, adult horses previously vaccinated bi-annually against EIV using the Fluvac Innovator vaccine (Zoetis). The horses were randomly assigned to one of three groups: 1- and 2-dose series with Fluvac Innovator, 1- and 2-dose series with Prestige II (Merck Animal Health) and 1- and 2-dose series with Vetera EIV (Boehringer Ingelheim). Five horses served as environmental sentinels. Whole blood was collected on the first day of vaccine administration and at 30, 60, 90 and 180 days. Serum samples were tested for antibody titers to EIV by hemagglutination inhibition assay against a contemporary EIV Clade I (Kentucky 2014) and Clade II (Richmond 2007) Florida sublineage strains. The geometric mean of the titers was compared amongst the EIV vaccine groups.

For all vaccine groups, there was a significant difference between antibody responses pre- and post-initial vaccine administration. There was no statistical difference after day 30 between any booster and single dose vaccine groups.

How does this research benefit horses? Equine influenza is one of the most devastating respiratory viruses. This study shows that, in previously immunized horses, a switch in vaccine manufacturer does not require a booster series in order to achieve superior antibody responses to Florida sublineage clade 1 and 2 EIV. This information will be relevant to educate veterinarians and owners on EIV vaccine protocols.

MEDICINE & INFECTIOUS DISEASE

Using microbiota analyses to understand and track a foal's gastrointestinal health (Grant #17-03)

Investigators: Michael J. Mienaltowski, DVM, PhD, Elizabeth A. Maga, PhD, K. Gary Magdesian, DVM, DACVIM, DACVECC, DACVCP, Ubaldo De La Torre

Foals need properly established gut microbiota to develop correctly and be healthy in their first years of life. Microorganisms associated with contact with the mare and consumption of colostrum and milk are the first to colonize a foal's GI system. As the foal's diet shifts from milk to solid food, so do the bacterial populations of the gut microbiota, enabling the foal to better obtain energy and nutrients from new feed sources. Disruptions to the microbiota, such as those that occur with diarrhea, can negatively affect foal health.

Fecal samples from foals were analyzed to detect differences in gut establishment by age, changes in diet, and diarrhea status. Data showed that the composition of bacterial populations in foals followed an age-dependent pattern linked to changes in diet. Differences in microbial population patterns

between healthy foals and those with diarrhea, as well as differences associated with management styles at individual facilities, were also observed.

Samples from foals during the first week of life showed a high abundance of Proteobacteria. From day seven through weaning, the study population shifted to an abundance of Firmicutes, the most abundant bacterial phylum seen in horses overall. As foals in the study were progressively exposed to solid foods, the abundance of Bacteroidetes also began to rise. Samples from foals with diarrhea showed an increased abundance of Enterobacteriaceae, a family of bacteria that includes *Salmonella* and *E. coli*.

How does this research benefit horses? Data from this study contributes to an improved understanding of the microbiota present in growing neonates and the role of the mare's milk in microbiota establishment. This could be used as a simple diagnostic tool utilizing fecal bacteria to provide veterinarians with information about foals' GI bacterial populations as they grow. A future goal would be for veterinarians to be able to suggest formulations of probiotics or screen for optimal gut microorganisms from healthy animals for transfer to sick animals. Further research could enable veterinarians to visit a farm that has horses with diarrhea and not just treat the horses, but combine genomics and epidemiology to troubleshoot a management issue on the farm to eliminate diarrhea, thus saving time, money, and improving equine welfare.

This research was reported in *PLoS One* 2019 April; 14(4): e0216211.

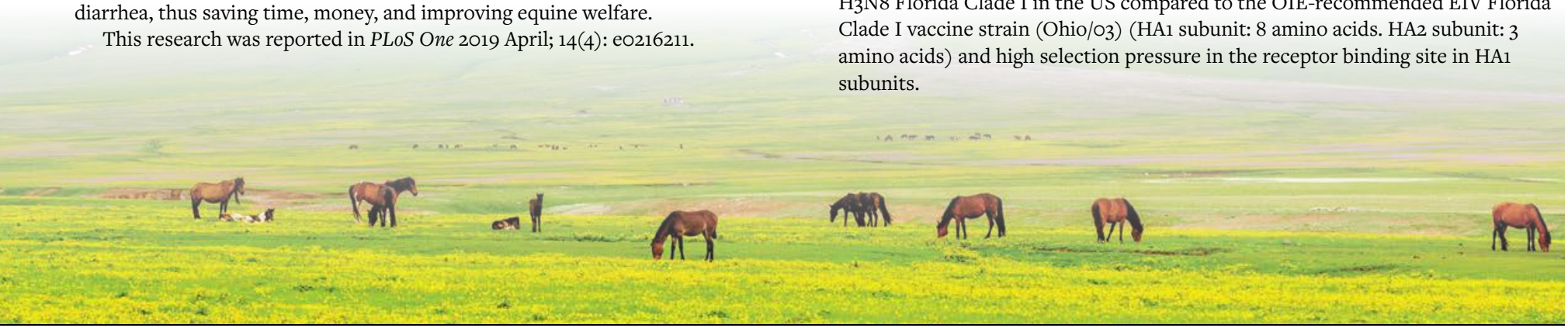
Investigating the epidemiology of equine influenza virus in the USA from 2006 to 2016 (17-10)

Investigators: Beatriz Martinez Lopez, DVM, MPVM, PhD, Nicola Pusterla, DVM, PhD, DACVIM, Samantha Barnum, MS, Kyuyoung Lee, DVM, MPVM

Equine influenza virus (EIV) is a highly contagious respiratory virus responsible for outbreaks of equine influenza (EI) worldwide. Today, only EIV of the Clade I and Clade II Florida sub-lineage circulate amongst equids. Since the virus continuously evolves and alters its morphology to escape the immune system of the host, it is important to determine how the structure of proteins responsible for the induction of a solid immunity change over time to best evaluate vaccine effectiveness and provide recommendations to ensure that contemporary vaccines contain epidemiologically relevant EIV strains.

This study investigated temporal and spatial molecular characteristics of H3N8 EIV isolates collected from horses in the USA from 2006 to 2016. Phylogenetic analysis showed that all current EIVs H3N8 were connected by two main trunk lineages, Florida Clade I and Florida Clade II. EIVs in Florida Clade I have endemically spread through North and South America, with a few spillovers into Japan, South Korea, Malaysia and Sweden. EIVs strains in Florida Clade II have mainly circulated among European countries and expanded to China, Mongolia and India. Among the study samples, 57 of the 58 EIVs were sub-lineages of Florida Clade I. A sample from a horse imported from Germany and quarantined in 2012 was a Florida Clade II virus.

Data showed 11 fixed amino acid substitutions in the HA gene of current EIVs H3N8 Florida Clade I in the US compared to the OIE-recommended EIV Florida Clade I vaccine strain (Ohio/03) (HA1 subunit: 8 amino acids. HA2 subunit: 3 amino acids) and high selection pressure in the receptor binding site in HA1 subunits.



How does this research benefit horses? Equine influenza is one of the most devastating respiratory viruses. This unique data set and analysis allow us to better understand why EIV cases in the US are becoming more prevalent today, even with the vaccine program combined with the implementation of quarantine measures for imported equids. Our study shows that introduction of novel strains of EIV or EIV clades 2 have not been observed in the US. However, continuous accumulation of fixed amino acid substitutions in the HA protein against the most recent EIV vaccine strains and their circulation could influence the effectiveness of commercially available EIV vaccines in the US. Updating EIV vaccine strains would help to improve the vaccine program against current EIVs in the US.

Investigation of the use of two different swabs for the detection of microorganisms in nasal secretions of healthy horses (17-13)

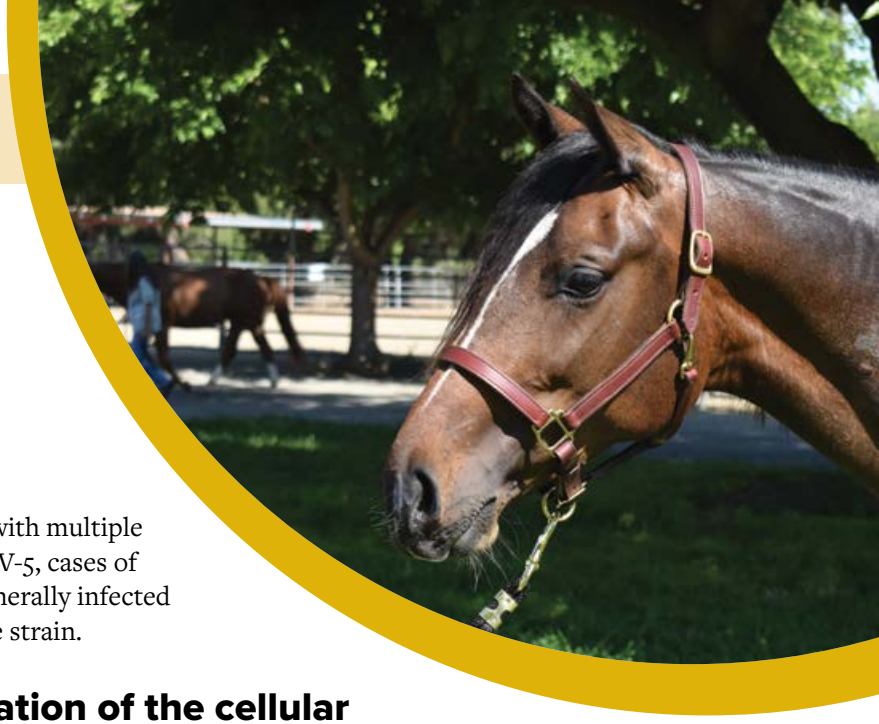
Investigators: Nicola Pusterla, DVM, PhD, DACVIM, DAVDC-Equine, Samantha Barnum, MS

Quantitative PCR (qPCR) is the diagnostic tool of choice for detection of respiratory microorganisms based on high sensitivity and specificity, quick turn-around-time, cost-effectiveness and panel testing. Given the clinical relevance of respiratory pathogens, it is important to use the best collection system that provides the highest detection yield in nasal secretions. While standard rayon-tipped swabs are routinely used for the collection of nasal secretions in horses, flocked swabs have demonstrated superior performance for the detection of human respiratory, vaginal and gastrointestinal pathogens.

The study objective was to determine if there is a difference in performance between flocked swabs and standard rayon swabs in detecting nasal carriage of ubiquitous viruses and bacteria among healthy adult horses. Thirty-one horses were swabbed with a flocked swab and rayon swab, processed for nucleic acid purification and assayed for target genes including the *eukaryotic equine glyceraldehyde-3-phosphate dehydrogenase* (*eGAPDH*) gene, the *glycoprotein B* (*gB*) gene of EHV-2 and EHV-5, the universal bacterial *16S rRNA* gene and the *ITS* gene of *Streptococcus equi* subspecies *zooepidemicus*.

Rayon swabs yielded significantly higher DNA and RNA concentrations than flocked swabs. There were no significant differences between the two swab types for the target genes. All swabs had detectable *eGAPDH* at the genomic DNA and complimentary DNA level and bacterial *16S rRNA* genes. Seven and 14 horses tested positive in both swabs for EHV-2 and EHV-5, respectively. An additional 11 horses tested positive only in one of the two swabs for EHV-2 or EHV-5. Five horses tested positive in both swabs for *S. zooepidemicus*, while an additional nine horses only tested positive in one of the two swabs.

How does this research benefit horses? Due to the economic impact of contagious respiratory pathogens, it is relevant that viruses and bacteria be detected with the most accurate and reliable protocol. Flocked swabs have been shown to be more accurate in the detection of human respiratory microorganisms. However, in this study, sampling with flocked swabs did not statistically improve the detection of selected target viruses and bacteria by qPCR. Some variability in the molecular detection of viruses and bacteria was observed between the two types of swabs collected from the same horse. The differences in rate of detection of microorganisms by qPCR between right and left nasal passages from the same horse highlight the importance of proper sampling and the need to standardize sampling procedures.



Investigating the genetic makeup of equine herpesvirus type 5 in healthy and sick horses

(Grant #17-15)

Investigators: Nicola Pusterla, DVM, PhD, DACVIM, DAVDC-Equine, Emir Hodzic, DVM, MSci, PhD

Equine herpesvirus-5 (EHV-5) is widespread in horse populations and is optimally adapted to its host, meaning that significant clinical expression of infection is rare. However, growing evidence supports the involvement of EHV-5 with upper airway infections and the more devastating equine multinodular pulmonary fibrosis (EMPF), a sporadic, progressive and fibrosing interstitial lung disease of adult horses. The objective of this study was to determine the genetic diversity of EHV-5 in biological samples of healthy horses, horses with upper airway disease and horses with EMPF.

Biological samples (nasal secretions, whole blood, bronchoalveolar lavage fluid and lung biopsies) were taken from 5 healthy horses, 5 horses with upper airway infection and 5 horses with EMPF. The samples were processed for next generation sequencing targeting the *glycoprotein B* (*gB*) gene of EHV-5. Genetic diversity of EHV-5 was determined for each sample type and compared amongst the various horse groups.

Healthy horses and horses with upper airway infection displayed a greater genetic diversity compared to horses with EMPF. Horses with EMPF had significantly less diversity in EHV-5 partial *gB* gene sequences (median 1) compared to healthy horses (median 8) and horses with upper airway infection (median 9) in nasal secretions. Healthy horses and horses with respiratory signs tested negative for EHV-5 in whole blood, bronchoalveolar lavage fluid and lung biopsies. However, individual horses with EMPF displayed similar EHV-5 strains independent of the biological sample type.

How does this research benefit horses? EHV-5 is a poorly characterized and fairly enigmatic gamma-herpesvirus. It is important to understand the viral dynamics of infections for EHV-5 in order to characterize the pathophysiology of this virus and develop better preventive and therapeutic protocols. Similar to human herpesvirus-4, the genetic diversity of EHV-5 appears to contribute to the clinical manifestation. While healthy horses and horses with respiratory signs are infected with multiple strains of EHV-5, cases of EMPF are generally infected with only one strain.

Investigation of the cellular response to *Sarcocystis neurona* in blood and cerebrospinal fluid of horses with and without equine protozoal myeloencephalitis (17-16)

Investigators: Nicola Pusterla, DVM, PhD, Diplomate ACVIM and AVDC-Equine, Patricia Conrad, DVM, PhD, Samantha Mapes, MS, Andrea Packham, MS

Equine protozoal myeloencephalitis (EPM) is a commonly diagnosed, economically costly neurologic condition, attributed to infection of the central nervous system with *Sarcocystis neurona* and/or *Neospora hughesi.* Laboratory diagnosis relies on the detection of antibodies specific to *S. neurona* and/or *N. hughesi* in serum and/or cerebrospinal fluid (CSF). Interferon-γ (*INF*-γ) is a cytokine involved in the host immune defense. This study aimed to evaluate the *INF*-γ response in horses with EPM.

The *INF*-γ gene expression in blood and CSF from horses with laboratory confirmation of EPM (serum/CSF ratio ≤ 64; n=18) was compared to the *IFN*-γ response of horses with no evidence of EPM infection (serum/CSF ratio > 64; n=24). *INF*-γ gene expression against the positive mitogen control and S. neurona was measured in almost all blood samples from horses with and without evidence of *S. neurona* infection. Due to the low number of nucleated cells in the CSF samples, only 3 and 5 CSF samples from horses with and without evidence of

S. neurona infection, respectively, had detectable *INF-γ* gene expression post-stimulation with the positive mitogen. The upregulation of *INF-γ* gene expression in blood from horses with and without EPM following stimulation with *S. neurona* was similar and ranged from 2.4-19.3 (median 7.3) and 3.0 to 22.4 (median 8.3), respectively. Despite the small number of CSF samples with detectable *INF-γ* gene expression following stimulation with the positive mitogen and *S. neurona,* there was no upregulation of *INF-γ* gene expression against *S. neurona* in five horses without EPM, while three horses with suspected EPM showed a 1.3-4.1 upregulation of *INF-γ* gene expression against *S. neurona.*

How does this research benefit horses? This study explored an untouched diagnostic field of EPM, which is the cellular response to *S. neurona* of mononuclear cells in blood and CSF from horses with and without EPM. While accurate diagnostic modalities are urgently needed in order to prevent overdiagnosing and underdiagnosing relevant infectious diseases, measuring cellular responses in the CSF of horses with EPM failed to consistently detect an upregulation of *INF-γ* gene expression secondary to *S. neurona.*

Development of a test that can identify the specific strain of multiple parasites thought to be associated with the neurological disease equine protozoal myeloencephalitis (Grant #17-17)

Investigators: Jeroen P. Saeij, MS, PhD, Nicola Pusterla, DVM, PhD, Diplomate ACVIM, AVDC-Equine, Patricia A. Conrad, DVM, PhD, Monica Aleman, DVM, PhD

Equine protozoal myeloencephalitis (EPM) is a debilitating neurologic disease associated with *Sarcocystis neurona,* and *Neospora hughesi* infection. Although many horses have antibodies against *S. neurona* (seroposivity), few develop EPM. Evidence suggests that co-infection with similar protozoan parasites, such as *Toxoplasma gondii,* is associated with a higher risk for developing EPM.

Diagnosis of EPM is challenging and relies on an indirect immunofluorescence antibody test (IFAT) to detect antibodies to the parasites. This project sought to develop an indirect ELISA test using immunogenic and polymorphic peptides that would distinguish antibodies in horse sera against *N. hughesi, N. caninum, S. neurona, S. fayeri,* and different *Toxoplasma* strains.

Serotyping is based on the exclusive reaction against the peptide from the genotype of the infecting parasite, but not against the corresponding peptide of other parasites. , Seventeen new candidate peptides from dense granule (GRA) secreted proteins, rhoptry (ROP) and surface related antigens (SRSs) proteins were selected, synthesized and tested on a panel of horse serum including EPM cases, healthy animals, and experimentally infected animals. High background and cross-reactivity were noted, even in the negative samples, likely due to the "sticky" nature of horse serum, making analysis difficult. Further testing was performed with the best serum samples that had antibodies against one or more of the parasites. Using a standard indirect ELISA protocol, the majority of the peptides did not react, except a few from the GRA6 protein. This could indicate that their antigenicity is not enough or that the technical conditions for the ELISA need to be improved.

How does this research benefit horses? Many horses are infected with *Sarcocystis neurona,* but only a small subset develop EPM. The ability to determine which parasite species/strains are present in ill horses is necessary to correlate infection specifics with disease outcomes and develop effective treatments. The information obtained from this study can be used to design serological tests to be able to identify the protozoan parasite/s that commonly infect horses and establish an etiological relation between EPM clinical signs and specific parasites.

ORTHOPEDICS & LAMENESS

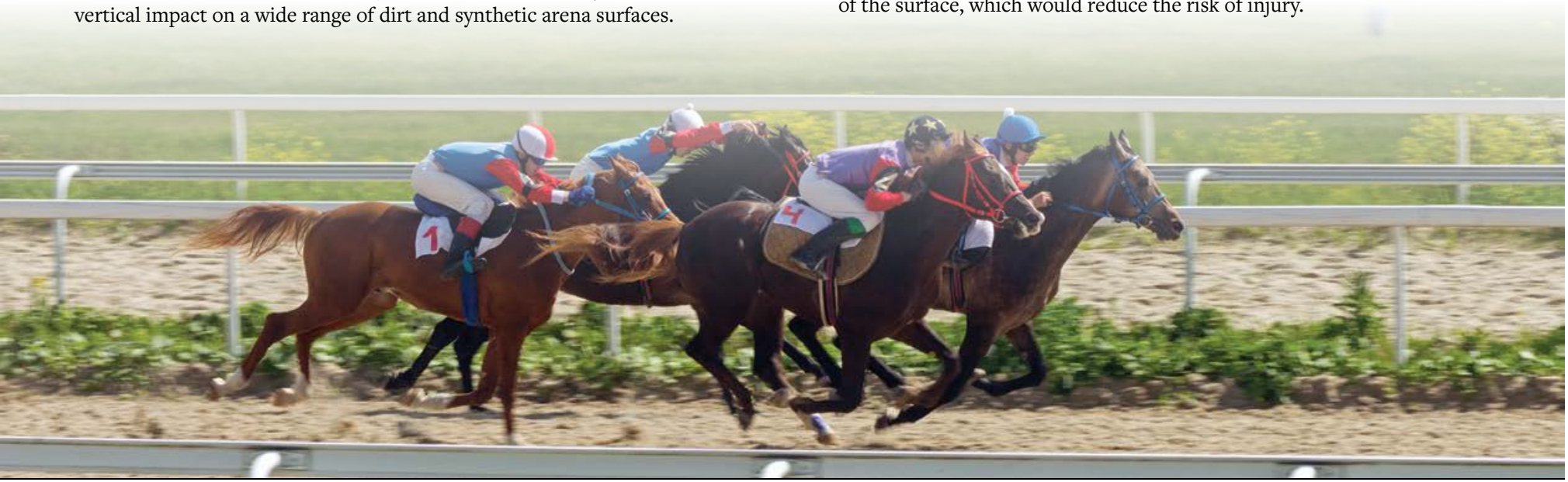
Determining the mechanical properties of different arena surface types (Grant #16-03)

Investigators: Susan Stover, DVM, PhD, DACVS, Christina Rohlf, BS, Tanya Garcia-Nolen, MS, Shrinivasa Upadhyaya, PhD, Sarah le Jeune, DVM, DACVS, CVA, DACVSMR, Cert Vet Chiro

This project was motivated by the need to understand the mechanical properties of dirt and synthetic arena surfaces in order to characterize the risk of injury to equine athletes performing on these surfaces. Previous studies within the laboratory indicated that synthetic surfaces exhibit a wide range of mechanical properties depending on factors such as surface preparation or composition, which may result in some synthetic surfaces having the same, or greater, injury risk than dirt surfaces. Although this laboratory has already developed validated equipment to test vertical impact via the arena impact device (AID), no validated equipment has been developed to measure shear surface properties. This study aimed to develop this shear testing equipment and quantify both shear and vertical impact on a wide range of dirt and synthetic arena surfaces.

The data showed that vertical ground reaction forces did not depend on surface type (dirt, synthetic) for freshly harrowed cushion. However, dirt surfaces exhibited higher vertical ground reaction forces (and may have a greater risk of injury) than synthetic surfaces for a previously trampled surface. Dirt surfaces exhibited higher surface compaction (cohesion) than synthetic surfaces and synthetic surfaces exhibited higher resistance to slide (angle of internal friction) than dirt surfaces. Synthetic surfaces exhibited higher cushion depth and moisture content than dirt surfaces, while dirt surfaces exhibited higher temperatures than synthetic surfaces. Correlations were found between surface management factors (cushion depth, moisture content, and temperature), shear, and vertical impact surface properties, indicating that surface mechanical properties can be adjusted by altering surface management protocols.

How does this research benefit horses? This research shows that dirt and synthetic arena surfaces vary in their mechanical properties. However, knowing that an arena has a dirt or synthetic surface is not enough information to understand how the surface behaves. Water, harrowing, and temperature affect the behavior of surfaces. Consistent surface properties would allow adaptation of the equine musculoskeletal system to the behavior of the surface, which would reduce the risk of injury.



SURFACE TESTING DEVICE.

How does horse forelimb motion during a jump relate to arena surface properties? (Grant #17-01)

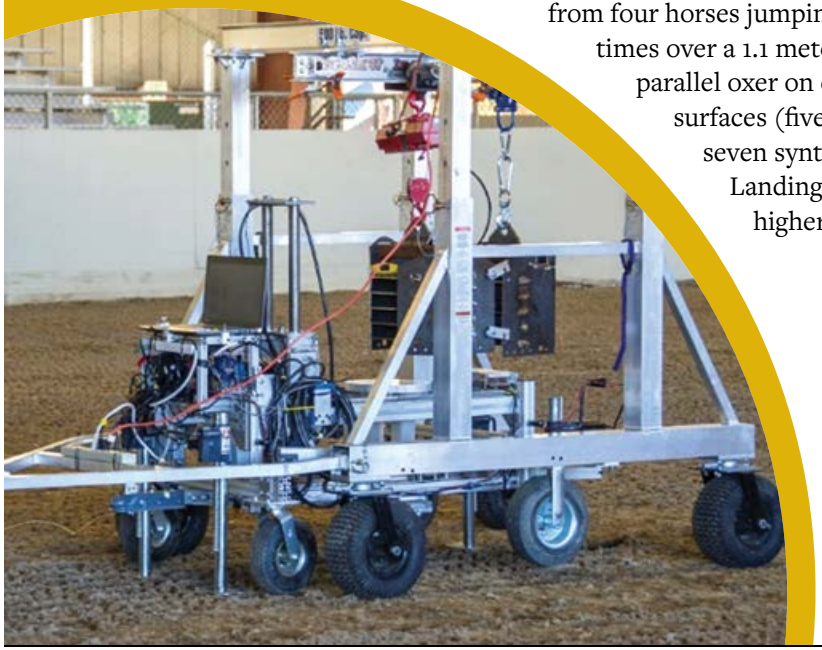
Investigators: Susan M. Stover, DVM, PhD, DACVS, Christina M. Rohlf, BS, Tanya C. Garcia-Nolen, BS, MS, Sarah S. le Jeune, DVM, DACVS, DACVSMR, CVA, Cert vet Chiro

Show jumping horses commonly injure tendons and ligaments in the lower limb, especially the suspensory ligament and superficial and deep digital flexor tendons as the result of large, repetitive loads that stretch the tendons and ligaments. The amount of stretching (strain) is affected by the amount and rate of fetlock joint extension. As footing can play an important role in fetlock joint extension, this study quantified relationships between surface properties and fetlock motion of show jumping horses to identify surface property variables that reduce the risk of injury.

Fetlock motion data was collected from four horses jumping three times over a 1.1 meter parallel oxer on each of 12 surfaces (five dirt and seven synthetic). Landing exhibited higher fetlock extension (4.8^0), rate of extension (668^0/s) and flexion (579^0/s), indicating that landing may put more strain on soft tissues that support the fetlock than takeoff. Fetlock angle was also higher when the instrumented limb was the leading leg (4.6^0), indicating that the leading leg may support more load and help stabilize the horse as the hindlimb lands. The risk of injury may be highest for the leading leg at jump landing.

Rate of fetlock flexion may represent the rebound energy available to the limb after contacting the surface. This was significantly higher in the dirt surface for takeoff (668^0/s) and landing (273^0/s), possibly because less energy is lost to soil movement. Rate of fetlock extension represents the extension rate of tendons crossing the fetlock joint and is maximized when hoof slide is reduced. High rates of fetlock extension may also lead to increased risk of injury.

How does this research benefit horses? Arena surface management is important. The 'feel' of an arena surface to the horse varies markedly with the depth of the cushion (harrowing) and water content (e.g., sprinkling). As soon as a freshly harrowed surface is impacted by hooves, the surface becomes harder.



◀ Surface testing device.

Effect of arena surface properties on hindlimb fetlock angle in show jumping horses (Grant #18-20)

Investigators: Susan M. Stover, DVM, PhD, DACVS, Lyndsey Marsh

The objective of this study was to determine if hindlimb fetlock motion during takeoff and landing from a jump differs between jumping on a dirt or a synthetic arena surface. There were no significant differences in fetlock motion between dirt and synthetic surfaces during take-off for the jump. The rate of fetlock extension was greater for dirt surfaces during landing after the jump. However, horse jumping style affected fetlock motion during take-off and landing. Leading limb and hoof position at surface contact reflect jumping style. Jumping style was relatively consistent within horses but varied among horses. When jumping style variability was considered, there were no statistical differences in hindlimb fetlock motion during takeoff and landing between jumping on a dirt or synthetic surface for the surfaces studied.

How does this research benefit horses? Lower limb injuries are prevalent in show jumping horses. While this research has suggested that there is not a significant difference between dirt and synthetic footing in the surfaces studied for hind limb fetlock motion in show jumping horses during takeoff and landing, it did provide insight that horse jumping style may play a significant role in fetlock motion. This study provides a window of opportunity into another avenue for injury prevention – training to influence jumping style.

The effect of horseshoe length on hoof wall changes that could lead to abnormal hoof conformation associated with injuries (Grant #16-13)

Investigators: Vanessa Dahl, MS, Susan M. Stover, DVM, PhD, DACVS, Tanya C. Garcia-Nolen, MS

Abnormal hoof conformation is related to muscle pain and lower limb lameness in performance horses, and fetlock injuries in racehorses. While horseshoes protect the hoof from excessive wear, they restrict other normal hoof functions. Therefore, shoeing techniques have the potential to affect hoof growth and cause abnormal hoof conformations, which in turn puts affected horses at risk for lameness and injury. We hypothesized that horseshoes mismatched in length to hoof size adversely affect hoof wall flexibility and cause hoof wall distortions that promote lameness and injury.

The effect of horseshoe length on hoof wall flexibility and distortion, and on fetlock extension (an index of fetlock injury), was investigated using cadaveric forelimbs from eight horses. Forelimbs were fitted with shoes of different lengths and loaded in a mechanical testing system to replicate maximum limb load at the canter. Each limb was tested 5 times to compare the following conditions: unshod, with a short shoe, full shoe, long shoe, and again unshod for comparison to initial conditions.

How does this research benefit horses? The data showed that fetlock extension decreased with the long shoe. In addition, application of a horseshoe to the hoof increased hoof wall strains and changed the way the hoof wall deformed. Horseshoes that extend to the back of the hoof (as opposed to shorter shoes) should reduce the likelihood of fetlock and flexor tendon injuries. Development of less rigid horseshoes may assist in preventing the development of abnormal hoof conformations.

The effect of horseshoe length on hoof growth, hoof horn tubule orientation and hoof wall angles (Grant #17-07)

Investigators: Vanessa E. Dahl, MS, Susan M. Stover, DVM, PhD, DACVS, Tanya C. Garcia-Nolen, MS, Ellen R. Singer, BA, DVM, DVSc, DACVS, DECVS, MRCVS, David A. Hawkins, PhD

Underrun heel hoof conformation is associated with muscle pain and lower limb lameness in performance horses, and fetlock injuries in racehorses. While horseshoes protect the hoof from excessive wear, they restrict other normal hoof functions, potentially affecting hoof growth and causing underrun heel hoof conformation. We hypothesized that horseshoes short in length relative to hoof size adversely affect hoof wall growth and cause hoof wall distortions that

promote underrun heel hoof conformation. The effect of horseshoe length on hoof wall growth and distortion was studied in 50 Thoroughbreds over six shoeing intervals, while shod with a normal length horseshoe for three intervals and a short length horseshoe for three intervals. Hoof wall angles, tubule orientation, and growth were measured before and after each shoeing interval.

Trends of decreasing toe angle and decreasing heel angle were found with a shorter shoe but seemed more associated with the time period (first half or second half), with no consistent results. Distal toe angles decreased for the short shoe on the left limb but increased for the right limb. No changes were noted for the angles along the quarter marks; however, a decrease in angle for the short shoe was found for the heels marks. Short shoe had a decrease in heel hoof length, quarter hoof length, and toe hoof length when compared to the full-length treatment.

Hoof conformation was not consistently different at the end of 18-week shoeing periods between horses shod with a short horseshoe compared with horses shod with a long horseshoe.

How does this research benefit horses? Racehorses with a long toe – underrun heel hoof conformation are at higher than normal risk for obtaining an unrecoverable fetlock injury. This study looked at one possible explanation for developing this abnormal hoof conformation. The length of the horseshoe does not appear to predispose sedentary horses to developing a long toe – underrun heel hoof conformation. Other factors, including exercise with differences in horseshoe length, should be examined.

How do horseshoe traction features alter hoof grip on performance footings? (Grant #18-25)

Investigators: Christina Rohlf, Tara Doherty, Susan M. Stover, DVM, PhD, DACVS

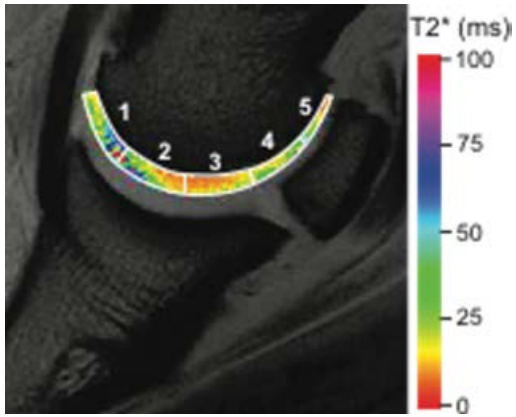
The grip of performance surfaces is a risk factor for lower leg injuries of sport horses by affecting the extent of hoof slide and stability of the leg. Horseshoe traction features may modify surface interactions. Surface grip was measured with eight paired cadaver hooves on a dirt surface (sand) and a synthetic surface (sand with fiber). Hooves were shod with positive (low toe grab), neutral (flat), and negative (sliding plate) traction characteristics. Unshod hooves served as a control. Surface material had a greater effect than horseshoe traction characteristics on surface grip with the synthetic surface exhibiting significantly higher grip than the dirt surface. Traction characteristics altered the shear force most notably on the synthetic surface with sliding plates exhibiting lower grip than the unshod hoof. However, traction characteristics did not affect the grip on the dirt surface.

How does this research benefit horses? The choice of surface material and design of horseshoe traction features to improve grip have previously been driven by subjective opinions. This study scientifically quantified the effects of surface material and horseshoe traction features on performance factors that directly affect the risk for leg injury in sport horses. Our results indicate that the addition of fiber alone to surface material can significantly alter the grip at the hoof-surface interface, while horseshoe traction features have a lesser effect on grip properties.

Improving visualization and assessment of cartilage in the horse foot (Grant #16-05)

Investigators: Derek D. Cissell, VMD, PhD, DACVR, Britton Nixon, DVM, Mathieu Spriet, DVM, MS, DACVR, DECVDI

Magnetic resonance imaging (MRI) is the gold standard imaging modality for non-invasive evaluation of joint injuries and damage to cartilage in people and animals. Our prior experience suggested that properties of cartilage in the joints of the horse foot cause it to appear different from cartilage in human joints. This research measured the



Color map demonstrating regional variation of cartilage properties in the coffin joint of the horse foot.

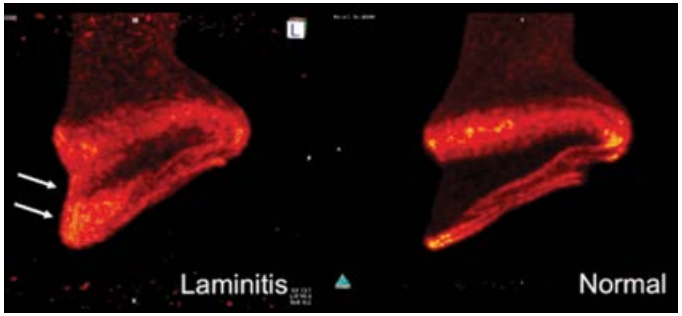
properties of cartilage in the horse foot that influence its appearance on MRI. We further sought to develop a novel MRI protocol to specifically visualize horse cartilage toward improved detection of cartilage damage. The properties of healthy cartilage in the horse foot differed significantly between different joint regions and also differed from reported values for healthy adult human cartilage. Based on our findings, we synthesized an artificial gel with MRI properties identical to horse cartilage. Using the gel, we tested twelve different MRI protocols and identified the best protocol for visualizing horse cartilage.

How does this research benefit horses? This study lead to the first MRI protocols developed specifically for imaging of cartilage in the horse foot. Protocols customized for MRI of the horse foot will improve visualization of cartilage in the foot toward better detection of cartilage changes associated with injury.

Use of Positron Emission Tomography as a new imaging modality for laminitis (16-09)

Investigators: Mathieu Spriet, DVM, MS, DACVR, DECVDI, Pablo Espinosa, DVM, Larry Galuppo, DVM, DACVS, Gary Magdesian, DVM, DACVIM

Laminitis is an extremely debilitating and often fatal disease in horses. Conventional imaging modalities cannot identify the early stages of the disease and provide only little information on the activity of the disease. We hypothesized that Positron Emission Tomography (PET), an advanced imaging modality recently applied to the horse at UC Davis, will identify early changes of laminitis and improve the assessment of the progression of the disease. In this study, we imaged seven normal horses and seven horses with laminitis with the PET scanner under general anesthesia. The results showed that the feet of all normal horses had the same appearance on PET whereas all feet with laminitis appeared different from normal feet. The most common abnormality in feet with active laminitis was evidence of changes in the front of the hoof wall, as expected. Feet with chronic laminitis showed a different appearance with normalization of the front of the hoof wall, but persistent changes in the coronary band, indicative of improper growth of the hoof.



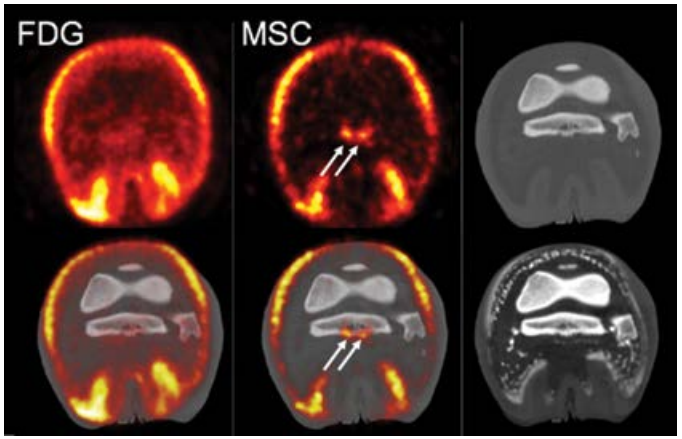
PET images of a foot with laminitis (left) compared with a normal foot (right). In this horse with a recent onset of laminitis, there is abnormal signal on the PET in the front of the hoof (arrows).

How does this research benefit horses? PET has the ability to distinguish different stages of the disease, which will help understand the progression of the disease and the response to different treatments. This will be extremely beneficial for development and assessment of new treatments for laminitis.

Stem cell tracking in the equine distal limb using Positron Emission Tomography (Grant #16-11)

Investigators: Mathieu Spriet, DVM, MS, DACVR, DECVDI

Assessing the fate of stem cells after their administration to a patient is critical to optimize treatments. Imaging techniques previously used in the horse for cell visualization provided only crude information regarding the localization of the cells. We hypothesized that Positron Emission Tomography (PET) will provide more



Horse foot imaged with regular PET (Left), PET for imaging of stem cells (middle) and CT (right). After injection of stem cells, two bright spots (arrows) appear at a site confirmed on the CT to be a tendon injury near the navicular bone. This indicates accumulation of stem cells at the tendon injury.

accurate information regarding cell localization in the horse foot. The objectives of this study were to assess the distribution of stem cells in the horse limb after injection and assess localization of stem cells in the foot of horses with tendon injuries. Four research horses, including two with tendon lesions, were injected in an artery of the limb, under general anesthesia, with stem cells labelled with a PET radiotracer. PET images of the limb were obtained at the time of injection and up to 2 hours later. A CT scan was also performed, and the PET and CT images were combined to assess the distribution of the stem cells.

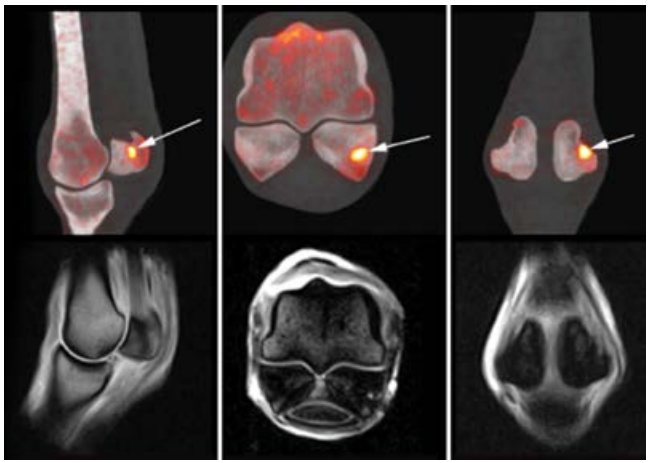
How does this research benefit horses? PET confirmed that the majority of the injected stem cells remained in the limb. Stem cells were identified at the site of the tendon injuries. Stem cells had a tendency to accumulate at the tendon injury. This study confirmed that the injection of stem cell in an artery of the limb leads to accumulation of stem cells at tendon injury. This is further evidence that this injection technique can be used to treat tendon injuries, which are a common occurrence in sport horses.

18F-Fluoride Positron Emission Tomography for detection of active osseous lesions in the equine distal limb. (Grant #16-15)

Investigators: Mathieu Spriet, DVM, MS, DACVR, DECVDI

Diagnostic imaging has made tremendous progress in identifying skeletal injuries in the horse over the past 15 years. However, early or subtle bone injuries remain difficult to detect. In addition, the significance of abnormalities identified with conventional imaging techniques is sometimes unknown. We hypothesized that Positron Emission Tomography (PET), an imaging technique only recently available to horses, will identify injuries not recognized using other imaging modalities and provide information regarding the significance of injuries. The objectives of this study were to validate the new imaging technique in research horses and apply it to clinical cases for which conventional imaging did not provide sufficient information to diagnose the cause of the lameness.

The results showed that PET imaging was well tolerated by all horses and provided high quality images. PET identified injuries not recognized with other imaging modalities. These include early injury in the fetlocks of racehorses (in particular in the sesamoid bones), early injury to the bone of joints including the hock, the fetlock, the pastern and coffin joints, early injury to the navicular bone and injuries at the attachment of the suspensory ligament on the cannon bone and of small ligaments in the foot. PET also helped distinguish between active and inactive injuries, which was particularly helpful in the hock.



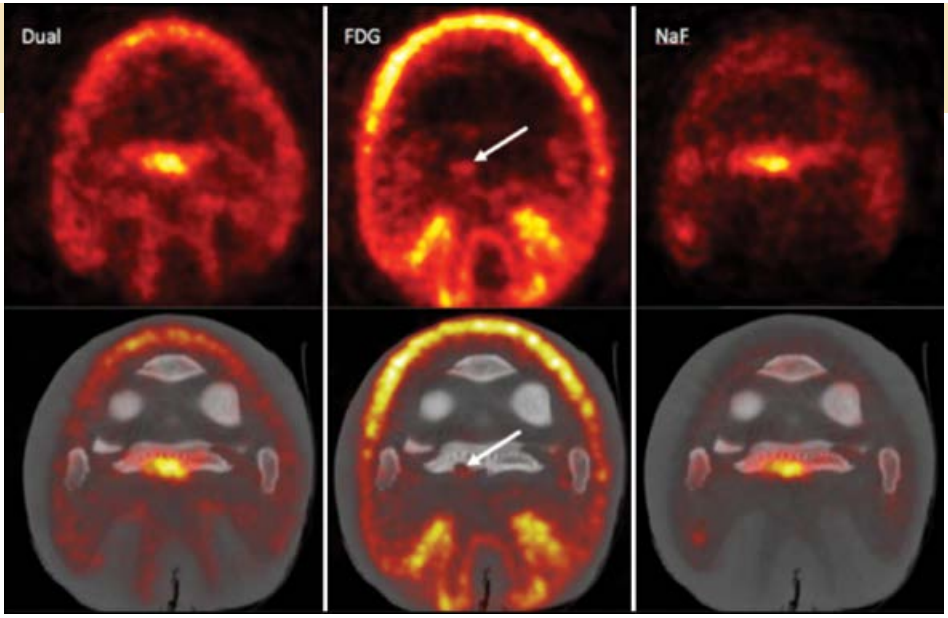
The PET (yellow area, arrows) demonstrates an early injury in the sesamoid bone of a racehorse. This was not detected on the MRI images (bottom row).

How does this research benefit horses? PET proved to be a valuable addition to other existing imaging techniques to assess injuries of bones in horses. This is of particular interest in racehorses, where PET can identify early bone changes before they progress to catastrophic injuries. PET is also very useful for the assessment of lameness in sport horses in the foot, fetlock and hock.

Development of a new technique for imaging bone and soft tissue of the horse limb (Grant #17-06)

Investigators: Mathieu Spriet, DVM, DACVR, DECVDI, Pablo Espinosa, DVM, Scott Katzman, DVM, DACVS, Larry Galuppo, DVM, DACVS

Positron Emission Tomography (PET) is an imaging modality that has recently become available to the horse. PET consists in administering a small amount of a special marker that will accumulate at the site of injuries and be detected by the scanner. There are different markers for soft tissue or bone injuries. So far, our team has validated the use of these markers used individually. However, in many horses, soft tissue and bone lesions coexist and it is important to recognize



PET images from a horse with navicular disease with combined markers (dual), soft tissue marker (FDG) and bone marker (NaF), top row, left to right. PET images are combined with CT images in the bottom row to provide anatomical localization. This case demonstrates that the combined scan easily recognized the injury of the navicular bone (yellow area in the center); however, a more subtle injury (arrow) in the adjacent tendon could not be recognized in the combined scan.

both. The goal of this study was to see if the markers could be combined within one scan. Six horses with soft tissue and bone injuries in the foot were scanned with the two tracers independently, and then combined. Overall, the scan with the combined tracers detected the majority of the injuries identified with the individual scans. The main exception was when a soft tissue injury was in close proximity to a bone injury and could be hidden by the bone injury. Almost all bone injuries could be recognized when the markers were combined. The combination of the tracers is advantageous as it decreases the amount of time a horse needs to be anesthetized to collect the information on both soft tissue and bone injuries. Although it is important to keep in mind the limitation identified, this study validated this technique for use in horses with lameness.

How does this research benefit horses? This study confirmed that combining two PET tracers in one scan could provide the information previously collected under two different scans. This technique is now commonly used at the UC Davis veterinary hospital, leading to better identification of causes of lameness in horses and helping to select treatment to improve healing and recovery.

Establishing the relationships between training and racing programs and bone damage and bone loss for the future prediction of fetlock fracture risk in Thoroughbred racehorses. (Grant #17-04)

Investigators: Susan M. Stover, DVM, PhD, DACVS, David P. Fyhrie, MS, PhD, Tanya Garcia-Nolen, MS, Sarah K. Schaffer, BS

Bone fractures result in removal of Thoroughbred racehorses from training and racing. When fractures are catastrophic, they often result in horse death and sometimes jockey injury. During racing, the fetlock sustains the highest loads of any part of the limb and often extends beyond the joint's physiological range of motion, making it extremely susceptible to injury. The most common fetlock injury is fracture of the proximal sesamoid bone (PSB). Evidence indicates PSB fractures in racehorses are repetitive overuse injuries. We used morphological microcomputed tomography (uCT) and histological samples to determine if grossly observed changes in the bone tissue of PSB fractures are correlated with the horse's training history.

Data showed that focal lesions exist in bones from the PSBs of racehorses euthanized due to unilateral biaxial PSB fracture. These focal lesions are not present in racehorses that died from other musculoskeletal injuries. There is a correlation between uCT morphological parameters and the horse's exercise history.

How does this research benefit horses? There are significant benefits from knowing the conditions that predispose racehorses to proximal sesamoid bone fracture, the most common unrecoverable fetlock injury. The study demonstrated that PSB fracture is the acute manifestation of a process that develops over time. It also documented the location, shape, and size of the pre-existing abnormality that predisposes horses to catastrophic PSB fracture. There are significant differences in how horses are trained and raced that separate horses that fracture PSBs from horses that do not fracture these bones. The data collected will be used to validate a finite element model that predicts internal changes in bone density and microdamage beneath the subchondral surface of PSBs in response to horse-specific training histories.

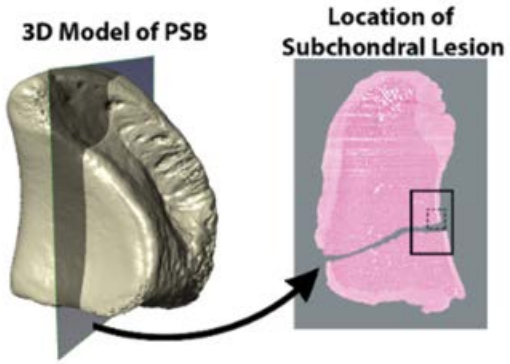
How are bones that fracture different from bones that don't fracture in racehorses with fetlock breakdown? (Grant #18-04)

Investigators: Sarah Shaffer, Susan M. Stover, DVM, PhD, DACVS, David P. Fyhrie, PhD

Identification of abnormalities in the fetlock bones of racehorses that died because of a fetlock breakdown can be used to understand why some horses have a fetlock breakdown. The proximal sesamoid bones (PSBs) are bones within the fetlock whose fracture cause 45-50% of racehorse fatalities due to injury.

Microscopic examination of fractured and non-fractured PSBs from racehorses revealed clusters of cracks that were most numerous in fractured bones, present but less numerous in non-fractured bones from racehorses that had another fetlock bone fractured, and rare in racehorses that died for reasons unrelated to fetlock breakdown. Cracks were in a consistent location in the bones. These cracks represent pre-existing microscopic damage that weaken bones and may predispose to bone fracture. There was also evidence of bone resorption to remove the microscopic cracks, which could further weaken bones until new bone has time to replace the damaged bone.

How does this research benefit horses? Fracture of the proximal sesamoid bones (PSB) is the most common fracture in Thoroughbred racehorses, causing approximately half of fetlock related fatalities. This study demonstrated that there are changes leading up to fracture that can be used to identify horses at risk for fracture – before they fracture. High-risk horses could be treated until the abnormalities heal, and then return to racing. Additionally, knowledge of the presence and consistent locations of these abnormalities can be used with advanced imaging, like PET scans, to detect horses at risk for fracture. Further, data from this study will be used to identify training and racing schedules that put a horse at high risk for fetlock breakdown.



This research was reported in the *Equine Veterinary Journal* 2020 May; 13291.

Determining the mean peak concentration of amikacin sulfate in the coffin joint during regional limb perfusion (Grant #16-10)

Investigators: Isabelle Kilcoyne MVB, DACVS, Jorge Nieto MVZ, PhD, Diplomate ACVS, ACVSMR, Julie Dechant DVM, DACVS, DACVECC, Heather Kynch DVM, PhD

Synovial sepsis of equine distal limb joints and traumatic wounds are a commonly encountered problem in equine practice. Regional limb perfusion allows the delivery of a high concentration of antibiotic to the affected region. Standardization and reduction of the tourniquet time required to perform these perfusions would allow treatment of these conditions in a more efficient manner.

The purpose of the study was to determine the most appropriate length of time for tourniquet application during distal limb perfusion with antibiotics in order to reduce pain and collateral side effects, as well as to increase the efficiency of this procedure. Seven healthy horses underwent intravenous regional limb perfusion (IVRLP) using standing sedation with 2g amikacin sulfate diluted to 60mls using 0.9% saline in the cephalic vein of a front limb using a pneumatic tourniquet placed 10 cm proximal to accessory carpal bone. Synovial fluid was collected from the coffin joint at 5, 10, 15, 20 and 30 minutes after IVRLP. The concentration of amikacin within the joint was measured at each time point. The median peak concentration (Cmax) of amikacin and the time to median peak concentration (Tmax) within the DIP joint was determined.

Results showed that the median peak amikacin concentration for the distal interphalangeal joint (DIP) joint was 550 (range 37-2167) µg/mL. The median time to peak concentration for the DIP joint was 15 minutes.

How does this research benefit horses? Traumatic wounds in the limbs of horses are common and frequently involve synovial structures that can affect the life and performance career of the animal. Distal limb perfusion with antibiotics is a simple procedure, which allows local administration of effective levels with minimal systemic effects. This study demonstrated that tourniquet application

of 15 minutes is sufficient for completion of intravenous regional limb perfusion when trying to achieve adequate synovial levels of amikacin in the coffin joint.

This research was reported in the *American Journal of Veterinary Research* 2018 Mar; 79(3): 282-286.

Unraveling the effect(s) of osteoporosis on bones and joints of the neck of horses suffering with neck stiffness and pain (Grant #16-19)

Investigators: Susan M. Stover, DVM, PhD, DACVS, Mathieu Spriet, DVM, Ms, DACVR, DECVDI, Sheley Nola, Brian Murphy, DVM, PhD, DACVP, Neil Willits, PhD

Neck stiffness and pain are common in horses that have osteoporosis and lung disease caused by breathing dust with toxic silicate particles. Affected horses may be unable to reach food on the ground and lose weight as a result. Increased understanding of the changes in the bones of the neck may lead to earlier diagnosis of horses with lung-associated osteoporosis and better management and preventive strategies to improve quality of life of affected horses.

We hypothesized that neck pain and stiffness in horses affected with osteoporosis can be attributed to bone degeneration and arthritis in the bones and joints of the neck. The necks of horses that died due to osteoporosis and of horses that died for reasons unrelated to osteoporosis were examined using computed tomography (3-dimensional radiographs). Abnormalities of the bones and joints were compared between affected and unaffected horses.

How does this research benefit horses? The reason for neck stiffness and pain in horses suffering from osteoporosis is not well understood, and thus, cannot be appropriately managed. Our results showed that silicate associated osteoporosis (osteoporosis due to inhalation of a toxic substance) affected bones of the neck, with the neck bones close to the chest being more severely affected. Degeneration of the bones also caused collapse of the intervertebral disks. Severely affected horses had marked neck stiffness and pain. Knowledge of these neck abnormalities in the neck allows for 1) early radiographic diagnosis of horses with osteoporosis, 2) early treatment of affected horses with medication, 3) appropriate management of affected horses (e.g., elevation of feed and water troughs), and 4) prevention of diagnosis of incorrect causes (e.g., wobbler) of neck pain in affected horses.

REGENERATIVE MEDICINE

Moon blindness: Defining immune cells and how stem cells may help decrease inflammation associated with moon blindness (Grant #16-16)

Investigators: Dori L. Borjesson, DVM, PhD, DACVP, Mary A. Lassaline, DVM, PhD, DACVO, MA, Rebecca R. Bellone, PhD, Naomi J. Walker, BS, Seldy G. Nelson, BS

Equine recurrent uveitis (ERU), or moon blindness, is the most common cause of equine blindness. It is caused by T lymphocyte-driven inflammation in the eyes (flares), and there is currently no cure. Mesenchymal stem cells (MSCs) are known to decrease T lymphocyte inflammation and may be an effective therapy for ERU. We hypothesized that horses with ERU have a specific "blood lymphocyte profile" that differs from unaffected horses, and that MSCs can switch specific lymphocytes from an inflammatory to a regulatory state.

Data showed that normal horses and horses with ERU had similar percentages of T and B lymphocyte subsets in blood. However in horses with ERU a subset of these T lymphocytes were shifted towards a pro-inflammatory response, they secreted significantly more interferon gamma (IFNγ) than T lymphocytes from normal horses. Many ERU horses also had T lymphocytes did not secrete as much of an anti-inflammatory cytokine, IL-10, as did normal horses. Lymphocytes from ERU horses also had high expression of a receptor that helps them move from blood to enter lymph nodes. This receptor means that the lymphocytes have been "primed" by previous exposure to an antigen. This is a common finding in animals with immune-mediated disease. These findings confirm that horses with ERU have a pattern of activated, pro-inflammatory lymphocytes that could serve as biomarkers of disease.

Expression of Foxp3 on CD8 T cells may be increased in ERU diseased horses. ERU horses tended to have increased expression of Foxp3 on CD8+ T lymphocytes compared to the control horses. There was no difference in Foxp3+ expression on CD4+ T lymphocytes between ERU horses and control horses.

How does this research benefit horses? ERU is a devastating immune-mediated disease that requires long-term medical management and can result in blindness. These initial data highlighted that CD4 T cells were the most interesting subset of cells altered by ERU. Overall, MSCs were seen to decrease the CD4+ T cell activation phenotype including, most notably, the ability to decrease CD4+ T cell IFNγ concentration, which is elevated in ERU horses. Stem cell treatment continues to be a promising indicator for treatment of CD4+ T cell mediated diseases in horses. This study also provides critical data in horses to compare to data in cats, dogs and humans, where MSC modulation of T cell subsets, including CD8+ cells, is increasingly recognized, and inform human clinical trials for patients with autoimmune uveitis for which the horse is an excellent model of disease. A deeper understanding of how MSCs work in the context of ERU will lead to a personalized medical approach to therapy.

REPRODUCTION

Variation in sperm mitochondrial function varies with stallion age and cryopreservation success and provides rationale for novel treatment and prevention strategies for male subfertility.

(Grant #16-07)

Investigators: Stuart Meyers, DVM, PhD, Gino Cortopassi, PhD, Evelyn Bulkeley, BS

Mitochondrial dysfunction has been implicated as a major factor in aging and age-related diseases in numerous species and tissue types. The objective of this study was to evaluate the relationship between stallion age, sperm quality, and mitochondrial function for age-related patterns of dysfunction in fresh stallion semen. This was accomplished by employing an array of mitochondrial oxidative phosphorylation inhibitors and uncouplers of mitochondrial oxygen consumption (MITOX) in fresh semen from Quarter Horse stallions. Results indicated a significant negative correlation between stallion age and both MITOX and spare respiratory capacity (SRC). No significant correlations were found between age and ejaculate volume, concentration, viability, sperm deformity index, percent morphologically normal sperm, or motility parameters, but significant, although weak, positive correlations were observed between Compα-t and stallion age, SRC, and sperm deformity index. Significant positive correlations were found between MITOX and both total and progressive motility. Increasing age resulted in a significant decrease in MITOX, while increases in progressive motility resulted in a significant increase in MITOX. A negative interaction of effects was observed between age and mitochondrial uncoupling, with each year increase in age resulting in a 6.2% reduction in SRC. Interestingly, a significant negative relationship was found between age and progressive motility during ETC inhibition. In the presence of ETC inhibition, every year increase beyond the age of 11 resulted in a 9.8% decrease in progressive motility.

How does this research benefit horses? These results confirm that aging is accompanied by decreased sperm mitochondrial function and indicate an age-related increase in dependence on mitochondrial oxidative function for progressive motility maintenance, strongly implicating mitochondrial dysfunction in the known age-related decrease in stallion fertility and sperm quality. There are detectable differences in mitochondrial efficiency that vary with stallion. Despite a decline in motility over time, oxygen consumption continues to increase since sperm are still viable and respiring even though they are becoming less motile. When we can powerfully predict the relationship of mitochondrial function to sperm function, effects from aging, and stallion fertility, we will be able to provide rational design of semen or male treatment to optimize stallion utilization. This could enable drug, stem cell, or immunologic-based therapies that may prevent or reverse oxidative injury, or other mechanisms that affect male fertility. Ultimately, this could allow more stallions to remain commercially viable for additional breeding seasons and this, in turn, will allow greater participation of stallions from various breeds in expanding international breeding programs.

Use of non-invasive embryo imaging to detect early embryonic developmental changes that predict the likelihood of later embryonic survival

(Grant #16-17)

Investigators: Bruce W. Christensen, DVM, MS, DACT, Stuart A. Meyers, DVM, PhD, DACT, Ghislaine A. Dujovne, DVM, MS, DACT

Early embryonic death is poorly understood in horse breeding. Higher rates of early embryonic death are noted when breeding older mares and performing advanced reproductive techniques like intracytoplasmic sperm injection (ICSI). Finding changes in early embryonic development that correlate with high or low embryo survivability would help target future research into causes of early embryonic death, and help with synchronizing recipient mares for ICSI procedures. We hypothesized that equine embryos have landmarks of developmental competence that are based on cell cycle and mitotic behavior, making possible the predictability of cultured embryos to progress to transferrable embryos.

We have established one pregnancy from an ICSI-derived embryo and successfully performed oocyte recovery a total of 52 times while successfully recovering immature oocytes that have been in vitro matured (IVM) and cultured prior to ICSI. In our initial experiments, 52% of IVM oocytes extruded a polar body and were injected with sperm collected the morning of ICSI, 30 hours post-TVA. A very low (<5%) blastocyst rate was observed. A second experiment was then performed using improved media. Culture conditions demonstrated marked improvement in maturation oocytes aspirated from mares and in successful ICSI, with a near 30% blastocyst rate, which is the industry standard. This demonstrated a significantly higher level of blastocyst development reflected by decreased timing from oocyte aspiration to *in vitro* culture, and from removal of any blood from oocyte culture medium. The results show a marked improvement in successful embryo development.

Further improvements in *in vitro* maturation, including follicular fluid and recombinant hormones, resulted in increased oocyte quality from aspirations and resulted in a blastocyst rate of 20%. One blastocyst from this cohort was transferred to a synchronized recipient mare and resulted in a positive pregnancy diagnosis, the first "all UC Davis" ICSI pregnancy.

How does this research benefit horses? Predicting how many post-ICSI embryos are going to mature out of a cohort, and when they will mature, would be a tremendous help in synchronizing recipient mares for fresh embryo transfers. Abnormalities associated with later pregnancy losses could be detected and choosing not to transfer those embryos could save time and money. Identifying stages in embryonic development when problems first occur will help focus future research into the causes of and possible solutions for preventing early embryonic death.

"Petri" the first "all UC Davis" ICSI foal.

SURGERY/ANESTHESIOLOGY

Pilot study: evaluating dosage and safety of a new pain drug in horses (Grant #16-02)

Investigators: Robert Brosnan, DVM, PhD, DACVAA, Claudia Sonder, DVM

Pain in horses is commonly managed with anti-inflammatory and opioid drugs. However, these drugs are not always effective and can produce undesirable side effects. We hypothesized that a new volatile analgesic discovered at UC Davis might exhibit analgesic effects in horses. This study examined dose ranges and safety of the analgesic by evaluating behavioral and biochemical effects of escalating doses in horses with spontaneously occurring orthopedic pain and lameness that was not fully ameliorated by conventional therapy.

Administration of this new volatile analgesic discovered at UC Davis produced improvement of pain for up to two hours for most doses studied. Horses also took significantly more steps in the four hours after drug administration than in the four hours prior to drug administration. It is possible that the drug allowed horses to be more active because it helped alleviate their pain. Additionally, we were able to collect expired breath samples from horses and measure drug concentrations in the lungs and extrapolate drug concentrations that were present in the blood. Adverse effects were deemed minimal as, except at the two highest drug doses administered, no horse exhibited evidence of ataxia or sedation. At all doses, horses had no statistical changes in complete blood counts (CBC) parameters or chemistry panels that were repeated over the course of 2 days. A future modification of this technique could allow veterinarians to measure the real-time concentration of this volatile analgesic in horses to help better direct drug therapy and dosing.

How does this research benefit horses? This pilot study has provided a dose range for a novel analgesic that does not cause sedation or toxicity in horses, but which may be associated with clinically relevant efficacy. These results support the need to conduct a larger, blinded, controlled analgesia study in horses that includes the highest no-observed-adverse-effect level dose of this new drug. Improved pain management will improve welfare of horses and potentially reduce the need for euthanasia of horses. New analgesics may also alleviate systemic complications that are associated with pain, such as decreased GI mobility, gastric ulceration, kidney insult, decreased wound healing strength, and possibly development of secondary laminitis.

GENETICS

Investigation of a region on horse chromosome 19 as the cause for bilateral corneal stromal loss in Friesian horses (Grant #17-24R)

Investigators: Kelly E. Knickelbein, VMD, Rebecca Bellone, PhD, Mary E. Lassaline, DVM, PhD

Bilateral Corneal Stromal Loss (BCSL), a disease resulting in progressive thinning of a small region of the cornea, is implicated as a genetic disorder in Friesian horses. Bilaterally symmetric thinning of the cornea can progress to permanent ocular damage and blindness if globe rupture occurs, which can be career or life ending.

A region on chromosome 19 previously identified as associated with BCSL was evaluated for differences in the DNA sequence between affected and unaffected horses. Fifty single nucleotide variants were identified in the region of interest. While none were perfectly concordant with the disease, the most associated variant was a missense variant in the *mucin 4* (*MUC4*) gene (c.778G>C, p.Val260Leu, P=1.04x10-4). This variant was present in 10 of 22 cases and found in only 4 of 54 controls. *Mucin 4* has been linked to a corneal disease in humans. Additionally, two deletions (a 242 base pair deletion in *NECTIN3* and an 809 base pair intergenic deletion) and one insertion (a 241 base pair insertion in *LSAMP*) were identified and genotyped in the same horses. None of these structural variants were perfectly concordant with disease phenotype, and only the intronic insertion in *LSAMP* was statistically associated with disease status (P=0.019). As such, these structural variants are not likely to be the cause of BCSL. Given the lack of identification of a variant perfectly concordant with the disease phenotype, it is likely that this is a complex disease with multiple genetic variants potentially contributing to the disease process.

How does this research benefit horses? Bilateral Corneal Stromal Loss in Friesian horses is a disease that typically requires emergency surgery to prevent rupture of the globe. If the disease progresses to globe rupture, loss of use of the horse is likely, making this an expensive and potentially devastating disease for horse owners. This work elucidated details about the inherited component of BCSL in Friesian horses and identified 53 variants for further investigation. The most associated variants are being further scrutinized for use in genetic testing for marker assisted selection and identification of at-risk horses for clinical evaluation.

IMMUNOLOGY

Investigation into the effect of common anti-inflammatory medications on the acute inflammatory marker serum amyloid A (Grant #17-22R)

Investigators: Callum G Donnelly, BVBiol, BVSc (Hons I), DACT, DACVIM, Nicola Pusterla, DVM, PhD, DACVIM, DAVDC, Sharon J Spier, DVM, PhD, DACVIM

Serum amyloid A (SAA), an acute phase protein, is a widely accepted monitoring tool for acute infectious or inflammatory diseases in horses, especially for sport horses traveling long distances for competition. These horses are also likely to be concurrently and/or chronically treated with anti-inflammatory drugs (NSAIDs) and antihistamines. The effect of NSAIDs and antihistamines administration on the acute inflammatory response in horses as measured by SAA is unknown and may reduce the diagnostic sensitivity of this early inflammatory marker.

Thirty horses of known *Corynebacterium pseudotuberculosis* exposure status (by titer) were pre-treated with flunixin meglumine (n=6), fircoxib (n=6) or diphenhydramine (n=6) prior to receiving a strong inflammatory stimulus – *Corynebacterium pseudotuberculosis* bacterin/toxoid vaccine. Two control groups included horses receiving the vaccine (and no drugs) (n=6) and those receiving no vaccine and no drugs (n=6). Horses were monitored daily over 14 days for the development of side effects Blood samples were collected every 24 hours for the first seven days following vaccination and then every 48 hours until day 14 post vaccination. SAA concentrations were determined using a commercial stall-side lateral flow immunoassay. Vaccination was repeated 30 days following the initial administration. Horses followed the same protocol of pre-treatment, observation and sampling as before.

Administration of flunixin meglumine, firocoxib or diphenhydramine did not reduce the acute inflammatory response as monitored by SAA. Booster vaccination resulted in a more pronounced inflammatory response compared to the initial vaccination, regardless of concurrent treatment.

How does this research benefit horses? Co-administration of NSAIDs or antihistamines at the time of vaccination does not result in a reduction in SAA and therefore does not appear to modify the acute inflammatory process induced by a strong antigenic vaccine.

This research is under review at the *Equine Veterinary Journal*.

MEDICINE & INFECTIOUS DISEASE

A new tool in the diagnosis of equine neurodegenerative diseases (Grant #17-20R)

Investigators: Lisa Edwards, DVM, DACVIM, Carrie J Finno, DVM, PhD, DACVIM

Diagnosis of neurologic disease in horses is challenging; investigation involves multiple diagnostics in addition to thorough clinical examination. For diseases such as equine neuroaxonal dystrophy (eNAD) and equine degenerative myeloencephalopathy (EDM), there is no definitive antemortem diagnostic available. Neurofilaments, structural proteins unique to the neuron, have been studied as biomarkers of neurologic disease in multiple species and show promising diagnostic utility.

Phosphorylated heavy protein (pNfH) has unique properties that make it useful as a biomarker of neurologic disease. We hypothesized that horses with neurologic disease (eNAD/EDM or cervical compressive vertebral myelopathy (CVCM)) have higher pNfH levels in blood and cerebrospinal fluid (CSF)

compared to normal horses, with eNAD/EDM affected horses having the highest pNfH levels. We hypothesized that blood and CSF pNfH levels in normal horses wouldl be < 2 ng/ml.

Blood and CSF pNfH levels in non-neurologic horses and neurologic horses diagnosed with eNAD/EDM or CVCM were determined using a species-validated enzyme linked immunosorbent assay (ELISA) kit.Blood and CSF pNfH was < 1.2 ng/ml in neurologically normal horses. Horses with blood pNfH of 1–2 ng/ml were 8.8 times more likely to have eNAD/EDM at necropsy. Horses with blood pNfH of > 2 ng/ml were 4.4 times more likely to have eNAD/EDM at necropsy. Horses with CSF pNfH > 2 ng/ml were 2.35 times more likely to have eNAD/EDM at necropsy. Serum appears to be more predictive of eNAD/EDM than CSF and may differentiate eNAD and CVCM affected horses.

How does this research benefit horses? Currently, there is no diagnostic test available for eNAD/EDM other than necropsy. Even at necropsy, the disease may go unrecognized due to the subtlety of the microscopic changes that occur in the nervous system. Measurement of blood pNfH can support the clinical diagnosis of eNAD/EDM without the need for euthanasia and necropsy evaluation. Elevated CSF pNfH levels can also document injury to the spinal cord, as seen in CVCM, and reduce the need for expensive and invasive diagnostics. Lastly, identification of eNAD/EDM affected horses via blood pNfH testing can facilitate selective breeding and targeted vitamin E supplementation in susceptible pregnant broodmares and foals. It is important to note that while increased pNfH levels provide useful prognostic information, normal blood or CSF pNfH does not rule out neurologic disease. This test is now available at UC Davis.

This research has been submitted to the *Equine Veterinary Journal*.

The Equine pNF-H, phosphorylated neurofilament ELISA test is now available through the Clinical Laboratory Services at the UC Davis veterinary hospital. This biomarker test represents the first diagnostic antemortem testing available for eNAD/EDM. It is recommended to submit both serum and cerebrospinal fluid (CSF) samples for the most accurate results. Please visit https://www.vetmed.ucdavis.edu/hospital/support-services/lab-services/clinical-laboratory-services/pnfh for more information.

Comparison of white and red blood cell counts in Warmblood and Thoroughbred horses (Grant #17-23R)

Investigators: Emily A. Schaefer, VMD, DACVIM, K. (Gary) Magdesian DVM, DACVIM, DACVECC, DACVCP, CVA, Judy Edman

Complete blood counts (CBCs) are routinely performed in adult horses to evaluate for clinical or subclinical inflammation and infection based on equine reference intervals. Clinically, we have noted that certain breeds of horses have CBC values that fall outside of established reference intervals and could be miscategorized as unhealthy based on these differences.

We hypothesized that warmbloods will have lower values for red cell parameters and white blood cell counts than Thoroughbreds and other breeds 3-19 years of age. We collected whole blood (10mL) from healthy horses and performed Complete Blood Counts at the hematology lab at the UC Davis

veterinary hospital. The horses were grouped by breed and CBC parameters compared between groups.

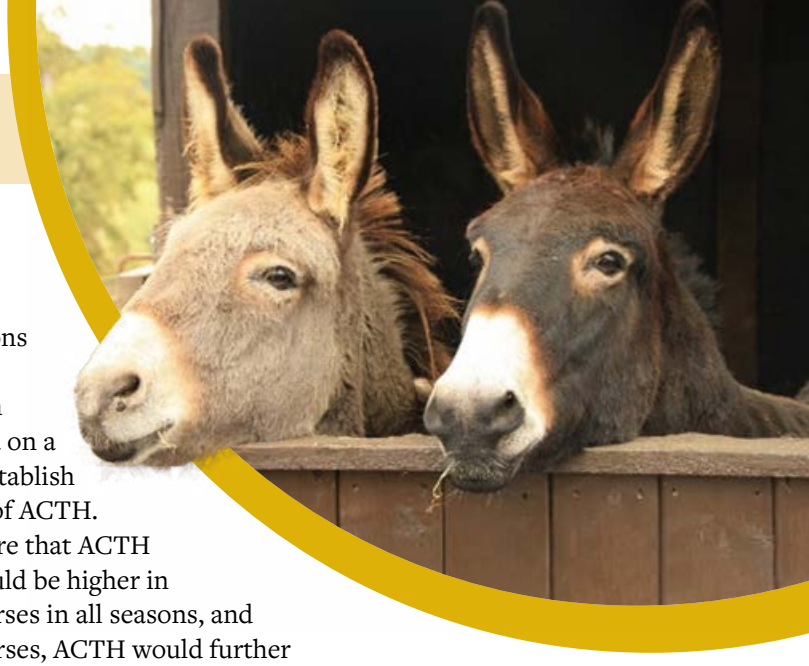
Warmblood horses had statistically significantly fewer total white cells compared to Thoroughbreds and other breeds. Warmbloods had statistically significantly fewer lymphocytes, a particular type of white blood cell. In fact, 26.5% of warmbloods had lymphocyte counts that were below the previously established lower reference interval. Warmbloods had statistically significantly lower red blood cell counts than Thoroughbreds. There were no statistically significant differences between breeds in eosinophil, basophil, or monocyte counts, nor fibrinogen concentration.

How does this research benefit horses? It has been previously documented that certain breeds of horses have differing numbers of certain types of blood cells. Thoroughbreds, for example, have higher red blood cell counts than many other breeds, a necessity for highly aerobic exercise such as racing as red blood cells carry oxygen throughout the body. This study documented for the first time that normal horses within a generalized subset of breeds (warmbloods) often have lower white blood cell counts than Thoroughbreds and other breeds. This is relevant in a clinical setting because it may alter treatment decisions when evaluating a warmblood horse with "low" white cell count and avoid unnecessary costs to client and delays in elective treatments for other conditions.

Seasonal variation of endogenous adrenocorticotropic hormone in healthy donkeys in Northern California (Grant #18-03R)

Investigators: Sarah Schale, DVM, Erin Goodrich, DVM, DACVPM, Philip Kass, DVM, MPVM, MS, PhD, Emily Berryhill, DVM, DACVIM

Pituitary pars intermedia dysfunction (PPID, or Equine Cushing's disease) is the most common endocrinopathy of aging equids and predisposes affected horses to significant health problems, including laminitis. A common test to diagnose PPID is assessing blood levels of adrenocorticotropic hormone (ACTH), which increases in horses with PPID. Seasonal variation of ACTH has been validated in normal horses to enable accurate year-round testing; however, these values have not been established in donkeys.



This study measured plasma ACTH concentrations in healthy, non-geriatric donkeys in northern California on a monthly basis to establish seasonal variation of ACTH. The hypotheses were that ACTH concentrations would be higher in donkeys than in horses in all seasons, and that similarly to horses, ACTH would further increase in the fall.

Twenty-five donkeys were recruited based on their age (median age 6 years, range 2 to 13 years) and appearance of good health based on physical examinations and complete blood counts. They were kept at the same property under the same management practices. Venipuncture was performed on a monthly basis from March 2019 through February 2020, and a validated ACTH assay was run on all samples. Months were grouped into seasons: spring (March through May), summer (June through August), fall (September through November) and winter (December through February).

Median ACTH concentration was 19.0 pg/mL in spring, 49.8 pg/mL in summer, 83.9 pg/mL in fall, and 12.6 pg/mL in winter. Each season's median ACTH concentration was significantly different from each other. Median ACTH concentrations fell within horse reference ranges in winter and spring months (< 35 pg/mL).

How does this research benefit horses? Based on the results in this group of donkeys, the best time to assess ACTH concentrations in donkeys is during the winter and spring months (December through May). This will avoid the large increases in ACTH concentrations that may occur in the summer and fall months. For winter and spring months it appears that horse reference ranges may be used. These findings will aid equine veterinarians in appropriate management of aging donkeys.

ORTHOPEDICS

Comparison of two advanced imaging modalities (PET/CT and MRI) for diagnosis of lameness localized to the foot in horses. (Grant #17-26R)

Investigators: Jannah Pye, BVSc, Mathieu Spriet, DVM, MS, DACVR, DECVDI

Computed tomography (CT) and magnetic resonance imaging (MRI) have considerably improved our ability to identify lameness-causing lesions in the equine distal limb, but the significance of some findings remains uncertain. Our group has demonstrated the value of Positron Emission Tomography (PET) in equine patients to assess bone and soft tissue lesions associated with lameness, but these should be compared to changes seen on MRI to better characterize the clinical relevance of such changes.

We hypothesized that dual PET/CT would allow assessment of both bone and soft tissue lesions detected on MRI in a clinical population of lame horses and detect lesions not visible on standing low field MRI. This study aimed to compare dual tracer PET/CT findings with MRI findings from horses that have lameness localized to the distal limb and correlate imaging results with lameness exam findings and outcome following treatment of identified lesions.

Horses (n = 8) that underwent both dual tracer PET/CT and MRI within the same month for evaluation of lameness localized to the distal limb by diagnostic anesthesia (palmar digital or abaxial sesamoid nerve block) were selected. The PET/CT findings were compared with the MRI findings and correlated with the patient's history, clinical findings, and outcome.

Results showed that PET/CT identified abnormalities not seen with MRI include deep digital flexor tendinopathy, desmitis of the chondrosesamoidean ligament and desmitis of the collateral ligament of the navicular bone. PET/CT also helped differentiate between "active" and "inactive" lesions in a horse with both navicular bone and deep digital flexor abnormalities seen on MRI.

How does this research benefit horses? PET/CT technique has the potential to improve detection of early or subtle injuries and help distinguish active from non-active lesions. With this modality now available at UC Davis, we anticipate that PET/CT will be a useful tool to help explain clinical findings and improve outcomes for horses with lameness localized to the foot.

SURGERY/ANESTHESIOLOGY

Can anesthesia be induced by intravenous etomidate in horses? (Grant #17-19R)

Investigators: Juhana M Honkavaara, DVM, PhD, Robert J Brosnan, DVM, PhD, DACVAA

Fewer anesthetic induction drug techniques exist for horses than for all other domestic species. The few available drug options can sometimes produce severe, undesirable cardiopulmonary effects in very ill horses. Etomidate is an injectable general anesthetic commonly used in humans and small animals that does not cause significant cardiovascular or respiratory depression at clinical doses. However, etomidate efficacy as a general anesthetic has never been tested in horses.

We hypothesized that etomidate can be used to induce general anesthesia in healthy horses. Sedated horses were administered etomidate in a stepwise manner based on the responses observed in the previous study horse ("Up-and-Down" method) to determine the median effective dose (i.e. the dose that will immobilize 50% of the study population) for general anesthesia. Clinically relevant physiologic responses (heart rate, respiratory rate, oxygen saturation, venous carbon dioxide), anesthetic depth signs (eye position, palpebral reflex, pupil size, limb or head motor activity), and induction and recovery quality were also be assessed.

Etomidate 0.5mg/kg administered to sedated horses was sufficient to produce recumbency and unconsciousness and allow intubation. However, induction quality in most horses was poor due to brief trembling and galloping behavior after anesthetic induction. Quality of anesthetic induction was not improved by higher etomidate doses or by addition of a benzodiazepine muscle relaxant. Although recoveries from etomidate alone were excellent, etomidate is an unsuitable anesthetic induction agent in horses.

How does this research benefit horses? Cardiovascular and respiratory depression can increase perianesthetic morbidity and mortality in sick horses (e.g., ill colic patients). General anesthesia that can be induced with minimal cardiopulmonary effect may provide a safer alternative to current agents in debilitated horses. However, based on the results of this study, etomidate is not suitable for general anesthesia in horses.

The efficacy of a 0.2% polyhexamethylene biguanide-impregnated gauze dressing against common orthopedic bacteria found in horses.

(Grant #17-25R)

Investigators: Charlene V. Noll BA, BE, MSME, DVM, Isabelle Kilcoyne, MVB, Diplomate ACVS, Jorge E. Nieto, MVZ, PhD, Diplomate ACVS & ACVSMR, Barbara A. Byrne DVM PhD DACVIM, DACVM

Traumatic wounds of distal limb joints are commonly encountered problems in equine practice, and frequently involve synovial structures. Such injuries can be career limiting or life threatening. Distal limb wounds in horses heal more slowly than wounds on other parts of the body because of a comparatively decreased blood supply, greater mobility over joints, and predisposition for bacterial contamination because of proximity to the ground. Common isolates from equine wounds include *Staphylococcus* spp. in addition to Enterobacteriaceae, *Streptococcal* spp., *Pseudomonas* spp., and anaerobes.

We hypothesized there would be significantly less bacterial growth of certain species of bacteria using the PHMB-impregnated gauze compared to the control gauze. Cultures of wounds, draining tracts and incisional infections were submitted for aerobic culture. Eleven aerobic bacteria were identified and banked at -20°C. Squares of PHMB-impregnated and non-impregnated control gauze were placed on Muller-Hinton agar plates inoculated with commonly isolated bacterial species (n=11). Growth under each gauze was assessed qualitatively after a 24-hour incubation period. Zones of inhibition were measured to a standardized scale, using image-processing software. A numerical scale was used to record level of inhibition of bacterial growth.

PHMB-impregnated gauze provided greater inhibition of growth of 4/6 Gram-positive species and 4/5 Gram-negative species on inoculated plates compared with control gauze. Growth inhibition (%) using the 0.2% PMHB-impregnated gauze for *Staphylococcal* spp. (n=4) ranged from 33-83.1% and for *Escherichia coli* spp. (n=4) ranged from 6.5-37% compared to 0% using the control gauze. There was no inhibition of growth of *Pseudomonas aeruginosa* or *Enterococcus* spp.

How does this research benefit horses? Use of a 0.2% polyhexamethylene biguanide (PHMB)-impregnated dressing resulted in variable growth inhibition of different *Staphylococcal* spp. and *Escherichia coli* spp, bacteria commonly encountered when treating orthopedic conditions such as infections post-operatively and wounds in horses. These dressings may be useful for reducing contamination of underlying wounds by common equine bacterial pathogens in clinical practice.

Arthroscopic surgery of the caudal cervical facets using a needle arthroscope (Grant #18-01R)

Investigators: Marcos Perez Nogues, LV, MSc, Betsy Vaughan, DVM, DACVSMR, Kathryn L. Phillips, DVM, DACVR, Larry D. Galuppo, DVM, DACVS

Caudal cervical osteoarthritis is a commonly found pathology that has a significant career impact in horses. The progression of the osteoarthritis in the lower neck can lead to neck pain, lameness, or subsequent spinal cord compression and ataxia, and can ultimately lead to retirement. Currently, the imaging modalities available usually have low yield correlating clinical signs with the severity of the lesions, and advance imaging is sometimes impossible

due to disparity of the equipment and horse´s size. In this study, we performed needle arthroscopy of the caudal cervical facet joint in healthy horses. The goal was to refine this technique in order to diagnose, prognosticate, and better guide treatment for osteoarthritic lesions, osteochondral (OCD) lesions and flush septic processes in these joints.

How does this research benefit horses? The study results showed that needle arthroscopic surgery of the caudal cervical facet joints can be done safely and successfully with the horses standing just under sedation. This minimal invasive surgery will be useful in the future for the diagnosis of neck pathology, could be used in future studies to discover how osteoarthritis impinge the spinal cord developing ataxia, and could be the start point of the developing of future treatments for neck osteoarthritis and other neck pathologies.

This research was reported in *Veterinary Surgery* 2020 Apr;49(3):463-471.

Determining whether or not the tarsocrural joint communicates with the talocalcaneal joint in the equine tarsus. (Grant #18-04R)

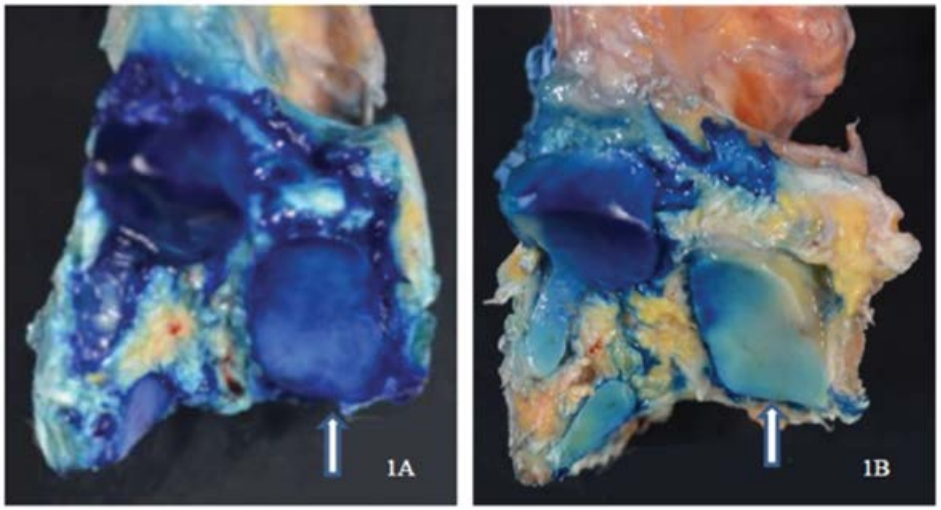
Investigators: Thomas Cullen BVMS, Katrijn Dow Whisenant DVM, Derek D. Cissell, VMD, Ph.D., DACVR, Larry D Galuppo, BS, DVM, DACVS

As the hock consists of four joints, communication between joints is an important aspect to consider when performing diagnostic or therapeutic joint injections. To date, the communication of the distal hock joints has been well investigated, but potential communication of the proximal joints remains unclear. If communication of the proximal joints exists, joint injection approaches could be altered to make injections more feasible in the field.

We hypothesized that injection into the front of the tarsocrural joint would result in diffusion to the talocalcaneal joint in the back of the hock. CT was performed on twelve paired limbs from horses euthanized for reasons unrelated to disease of the tarsus. The tarsocrural joints were injected with a mixture of iodinated contrast and methylene blue stain via a routine dorsomedial approach. One joint was injected with 60 mls (high volume) and the other was injected with 7 mls (low volume). Repeat CT following injection and flexion of the tarsus was performed. The tarsi were subsequently dissected to assess the methylene blue staining of the talocalcaneal joint cartilage.

With CT, contrast was noted to fill the tarsal sinus and surround the facets of the talocalcaneal joint in all 12 limbs. Discernable contrast between the articular surfaces of the talocalcaneal facets was only seen in 3/12 limbs. Methylene blue staining of all talocalcaneal facets was present in 12/12 limbs. For the high-volume limbs, the stain uptake was subjectively increased when compared with the low-volume injectate. The medial talocalcaneal facet had consistently less stain uptake than the lateral facets.

How does this research benefit horses? Methylene blue stain confirmed tarsocrural-talocalcaneal joint communication in all cases and was far superior to contrast-enhanced CT for this determination. Clinically, the majority of talocalcaneal joint disease is concentrated at the medial facet of the joint. Knowing that, in healthy limbs, there is limited communication with the tarsocrural joint into the medial facet decreases the likelihood that intraarticular treatments injected into the tarsocrural joint will be efficacious in clinical cases.



Dissected Calcanei after high volume (A) and low volume (B) injection with methylene blue stain via a dorsomedial approach to the tarsocrural joint. Lateral is to the left in both images. The medial facet (white arrows) is less stained than the lateral facets in both but markedly less in the low volume injection.

PARTNERSHIPS LEAD TO INNOVATIONS IN VETERINARY CARE

One of the many strengths of the UC Davis School of Veterinary Medicine is the guiding principle of collaboration in a multi-disciplinary approach to solve complex problems. These partnerships combine to investigate disease, improve techniques, identify treatments and advance knowledge.

Claire Giannini Hoffman Equine Athletic Performance Laboratory – Capabilities in equine sports medicine are enhanced significantly with the Claire Giannini Hoffman Equine Athletic Performance Laboratory (EAPL). This state-of-the-art, climate-controlled facility includes two high-speed Mustang treadmills, a video motion analysis system, and the laboratory equipment and support necessary to perform in-depth investigations of respiratory, cardiac, musculoskeletal, and metabolic causes of poor performance and exercise intolerance. The EAPL is home to an integrated multidisciplinary clinical and research equine sports medicine program anchored by Dr. Jim Jones, an internationally-renowned equine exercise physiologist, and supported by faculty from the veterinary hospital's Equine Surgery and Lameness, Equine Ultrasound, and Equine Medicine Services.

J.D. Wheat Veterinary Orthopedic Research Laboratory – The J.D. Wheat Veterinary Orthopedic Research Laboratory is an environment in which multidisciplinary studies pertaining to musculoskeletal disorders of animals and humans can be conducted. The goal of researchers participating in the laboratory is to understand the physiologic process of injury and musculoskeletal disease in performance, companion and production animals as well as in humans.

Kenneth L. Maddy Equine Analytical Chemistry Laboratory – The Kenneth L. Maddy Equine Analytical Chemistry Laboratory provides a drug testing program with the highest quality standards, employing the most innovative methodology and newest analytical technology, in order to ensure the integrity of horse racing. The Laboratory's two-fold mission includes expanding and disseminating new information regarding therapeutic medications in order to improve the welfare of California performance horses.

Veterinary Center for Clinical Trials – The Veterinary Center for Clinical Trials (VCCT) is advancing medical care for horses by developing and investigating alternative diagnostic approaches for a variety of diseases. The VCCT is frequently enrolling equine patients for a variety of studies, ranging from cardiology to orthopedics.

Veterinary Genetics Laboratory – The Veterinary Genetics Laboratory (VGL) provides animal parentage verification, identification, forensics services, genetic diagnostics and genetic disease research as a self-supporting unit of the UC Davis School of Veterinary Medicine. The laboratory is internationally recognized as a pioneer and expert in DNA-based animal testing. VGL also offers an extensive animal forensic services program, diagnostic tests for genetic diseases, and support for genetic research in domestic species, primates and wildlife.

Veterinary Institute for Regenerative Cures – The UC Davis School of Veterinary Medicine is a national leader for veterinary regenerative medicine under the direction of the Veterinary Institute for Regenerative Cures. The Institute has established laboratory techniques and animal models that have been used to study regenerative therapies for veterinary and human medicine. It has characterized equine stem cells isolated from different tissues (i.e. fat, bone marrow, umbilical cord blood and umbilical cord tissue) with a focus on adult-derived mesenchymal stem cells. The Institute has a foundation in collaborative, interdisciplinary "disease teams" that include basic research faculty and clinical faculty that focus on "bench to bedside" translation of stem cell therapies.

William R. Pritchard Veterinary Medical Teaching Hospital – The William R. Pritchard Veterinary Medical Teaching Hospital provides cutting edge equine care by board-certified experts in equine medicine and surgery at the most advanced and comprehensive veterinary hospital in the world.

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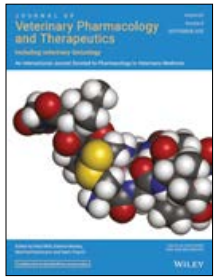

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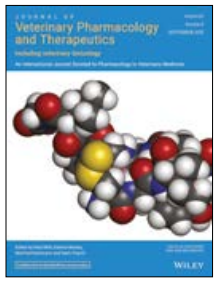

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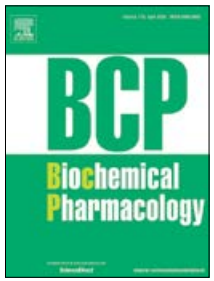

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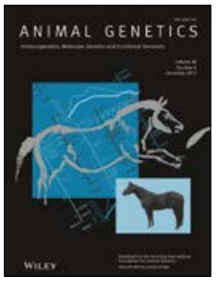

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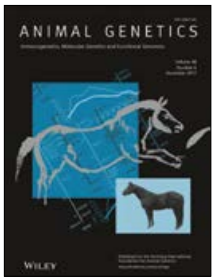

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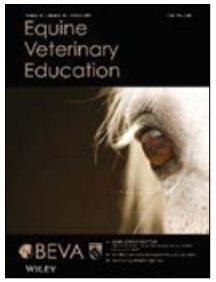

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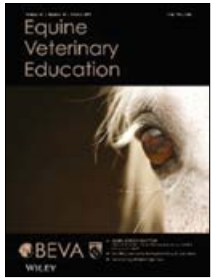

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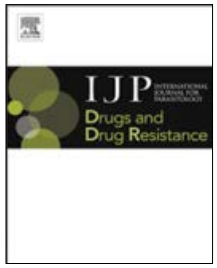

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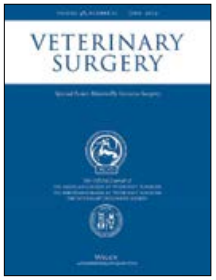

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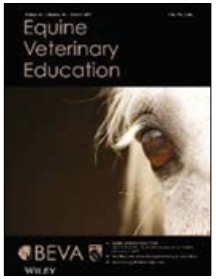

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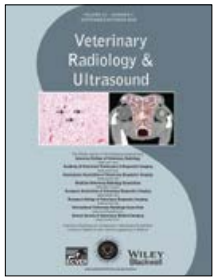

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Jon Kelly

In Tribute

The Center for Equine Health extends a special tribute to Mr. Ron Malone and Mr. Jon Kelly. These lifelong horsemen were devoted supporters of equine research through CEH. Their contributions not only helped to advance equine veterinary medicine, but also were magnified through the training and education of students and residents that now have successful careers in veterinary medicine. We are grateful for their dedication to these incredible animals that bring happiness to so many.

Ron Malone

NEWLY FUNDED RESEARCH STUDIES

- Assessment of clinical laminitis progression with the use of standing 18F-FDG positron emission tomography
- Baseline systemic parameters after transvaginal aspiration of oocytes in mares
- Characterization of a novel opioid analgesic in horses
- Comparison of prenatal ultrasonography and postnatal radiography for assessment of skeletal maturation in foals
- Comparison of standing MRI and standing 18F-NaF PET in horses with foot lameness
- Genomic characterization of ECoV strains from foals and adult horses
- Impact of arena surface properties on hindlimb motion and hoof translation of show jumping horses
- Inhibition of glycolysis to promote tendon formation
- Investigating the relationship of forelimb tendon and ligament strains and arena surface properties during jumping
- Mitotherapy for osteoarthritis (OA) related synovitis treatment – a new tool for veterinary medicine
- The effects of positive end-expiratory pressure on alveolar tidal recruitment/derecruitment and overdistention, gas exchange and cardiovascular function in horses anesthetized in dorsal recumbency

- The role of critical factors in equine egg maturation using time-lapse microscopy (TLM)
- Predicting the 1st cell division of equine embryos using time-lapse imaging
- Validation of CAPN9 as a risk haplotype for the development of melanoma in graying Connemara ponies
- Validating a genetic test for atypical equine thrombasthenia
- Validation of phosphorylated neurofilament heavy chain as an antemortem diagnostic for NAD
- Validation of a putative genetic mutation for inherited hypocalcemia in Thoroughbred foals
- Validation of two multiplex real-time PCR assays based on single nucleotide polymorphisms in order to differentiate between field and vaccine strains of *Streptococcus equi* subspecies *equi*

CENTER FOR EQUINE HEALTH RESEARCHERS

Monica Aleman, MVZ, Ph.D., DACVIM

Dr. Monica Aleman obtained her veterinary degree at the University UNAM-Mexico. She completed residencies in large animal internal medicine (equine emphasis) and neurology and neurosurgery at UC Davis and achieved board certification for both specialties by the American College of Veterinary Internal Medicine. She completed a Ph.D. in comparative pathology of neuromuscular diseases at UC Davis. Her research and clinical interest has focused in neurology, neuromuscular and muscle disorders in all species, with an equine emphasis. Dr. Aleman is a faculty member in the equine internal medicine and neurology services, chief of the equine internal medicine service, and Co-Director of the Neuromuscular Disease Laboratory at UC Davis. She is one of the founding members of the Equine and Comparative Neurology Research Group, and is affiliated with the Clinical Neurophysiology Laboratory at UC Davis.

Danika Bannasch, DVM, Ph.D.

Dr. Danika Bannasch earned her veterinary degree from the UC Davis School of Veterinary Medicine and her Ph.D. degree in mouse molecular genetics at Princeton University. She is currently a professor in the Department of Population Health and Reproduction in the UC Davis School of Veterinary Medicine and is the first faculty member to hold the prestigious Maxine Adler Endowed Chair in Genetics. An accomplished veterinary geneticist, Dr. Bannasch focuses her research on the identification of the molecular causes of inherited diseases in dogs and horses. Her laboratory has identified the DNA changes responsible for Lethal White Foal Syndrome, Hereditary Equine Regional Dermal Asthenia, and more. Important research findings have also led to animal models used for similar human diseases. By studying naturally occurring diseases in animals, the Bannasch Laboratory is discovering a triad of significant advances: the development of diagnostic tests to aid animal breeders; the identification of novel genes and pathways as candidates for human disease; and an understanding of basic molecular mechanisms of disease.

Rebecca Bellone, Ph.D.

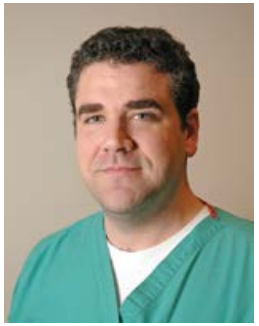
Dr. Rebecca Bellone earned her Ph.D. in Equine Genetics from the University of Kentucky in 2001. Subsequently, she has led an equine genetics research program involving both graduate and undergraduate students investigating the genetics of pigmentation and ocular disorders and the connection between the two. Her research team has collaboratively discovered causative mutations for both congenital stationary night blindness and ocular squamous cell carcinoma in horses. She is currently an Associate Adjunct Professor in the Department of Population Health and Reproduction and is the Director of the Veterinary Genetics Laboratory, a unit of the UC Davis School of Veterinary Medicine with an international reputation as experts in veterinary genetic testing.

Emily Berryhill, DVM, DACVIM

Dr. Emily Berryhill obtained her veterinary degree from the University of California, Davis, School of Veterinary Medicine in 2010. She completed the Large Animal Internal Medicine Residency at the University of California, Davis, School of Veterinary Medicine in 2016 and obtained Diplomate of the American College of Veterinary Internal Medicine status in 2016. She is an assistant professor in the Department of Medicine & Epidemiology. Dr. Berryhill is a faculty clinician in the Equine Internal Medicine Service. Her research focus is on equine physiology, endocrinology, and oncology, with a specialty focus on equine internal medicine. Additionally, she has performed pharmacologic studies evaluating new medications in horses.

Dori Borjesson, DVM, MPVM, Ph.D.

Dr. Dori Borjesson earned her DVM and MPVM degree from the UC Davis School of Veterinary Medicine in 1995 and completed a residency at UC Davis in Clinical Pathology in 1999 followed by a Ph.D. in Comparative Pathology at the UC Davis Center for Comparative Medicine in 2002. She joined the faculty as an assistant professor at the University of Minnesota for four years before returning to UC Davis as an associate professor in 2006. She serves as a professor in the Department of Pathology, Microbiology and Immunology and is the director of the Veterinary Institute of Regenerative Cures. Her research focuses on mesenchymal stem cells and immunomodulation. Her team works to define and develop naturally occurring animal models of disease to test cell therapies to improve animal health and inform human medical practice.



Robert Brosnan, DVM, Ph.D., DACVAA

Dr. Robert Brosnan earned his veterinary degree from the UC Davis School of Veterinary Medicine in 1999, and a Ph.D. in Physiology from UC Davis in 2002. He is a diplomate of the American College of Veterinary Anesthesia and Analgesia. Dr. Brosnan has developed technology that has identified agents in several novel classes that could lead to better, safer and more cost effective general anesthetics for use in operating rooms and surgical centers. His research focuses on cardiovascular and respiratory effects of anesthetics and on the mechanisms of anesthetic action. Dr. Brosnan is currently a professor in the Department of Surgical and Radiological Sciences.

Jennifer Cassano, DVM, Ph.D.

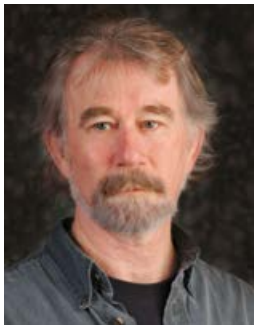
Dr. Jennifer Cassano joined the Equine Field Service as an assistant professor in 2019. Dr. Cassano received her DVM (2013) and PhD (Comparative Biomedical Sciences, 2016) from Cornell University. Upon completion of graduate school, she completed a combined academic/private practice one-year rotating internship (2017) at the Cummings School of Veterinary Medicine, Tufts University/Massachusetts Equine Clinic. She then worked as an associate veterinarian at EquidDoc Veterinary Services in Massachusetts. Her research interests and expertise are in the general area of stem cell biology and therapeutic actions of mesenchymal stem cells (MSCs), particularly in alterations in gene expression profiles of MSCs during exposure to inflammatory environments, and in the use of licensing agents to create more uniform MSCs exhibiting therapeutic traits such as chondroprotective activity.

Alessia Cenani, DVM, DACVAA

Dr. Alessai Cenani is an assistant professor in the Department of Surgical & Radiological Sciences. She received her veterinary degree from the University of Perugia, Perigia, Italy in 2009 and a Master's degree from the University of Liege, Liege, Belgium in 2012. Dr. Cenani came to UC Davis in 2016 for an anesthesia residency and subsequently became a diplomate of the American College of Veterinary Anesthesia and Analgesia. Her research focus is on pain management and recognition, as well as mechanisms of action of anesthetic and analgesic drugs, both in vitro and in vivo, with particular emphasis on assessment of drug efficacy in veterinary species.

Derek Cissell, VMD, Ph.D., DACVR

Dr. Derek Cissell earned his veterinary degree from the University of Pennsylvania in 2005. Following veterinary school, he worked for two years in a private, mixed-animal practice in northern Utah before returning to academia. He completed a residency at the UC Davis School of Veterinary Medicine in 2011, as well as earning his Ph.D. in biomedical engineering from UC Davis in 2015. Dr. Cissell joined the faculty in 2016 as an assistant professor in diagnostic imaging. His clinical and research interests include large animal diagnostic imaging and early diagnosis of cartilage injuries.



Alan Conley, BVSc, MS, Ph.D., FRCVS

Dr. Alan Conley is a professor in the Department of Population Health & Reproduction, Director of the Clinical Endocrinology Laboratory, and holds the John P. Hughes Endowed Chair in Equine Reproduction. His veterinary degree was awarded by the University of Melbourne and he saw dairy practice and mixed practice in Australia and in Scotland before completing a residency in theriogenology, and then Masters and Ph.D. degrees at Iowa State University. He was an NIH Fellow at UT Southwestern Medical Center in Dallas, a Research Scientist with the USDA in Nebraska and on faculty at North Dakota State University before coming to UC Davis. He earned a Diploma of Fellowship from the Royal College of Veterinary Surgeons (FRCVS) in recognition of his contributions to comparative reproductive physiology. Much of this work has related to sex steroid synthesis, but in recent years with a particular focus on equine reproductive endocrinology and developing new diagnostic endocrine assays.

Julie Dechant, DVM, MS, DACVS, DAVECC

Dr. Julie Dechant received her DVM from the University of Saskatchewan in 1996 and completed an MS and surgical residency in 2000 at Colorado State University. After faculty appointments at Saskatchewan and Oklahoma State University, Dr. Dechant joined the UC Davis School of Veterinary Medicine faculty in 2004 and is currently a professor in the Department of Surgical and Radiological Sciences and chief of the equine emergency surgery and critical care service. Dr. Dechant is a diplomate of the American College of Veterinary Surgeons and the American College of Veterinary Emergency and Critical Care. In 2014, she was elected a Fellow in the Teaching Academy of the Consortium of West Region Colleges of Veterinary Medicine.

Ghislane Dujovne, DVM, MS, DACT

Dr. Ghislaine Dujovne obtained her DVM from the University of Chile, College of Veterinary Sciences in 2004, followed by her Diploma in Animal Reproduction with an equine emphasis. She worked in private general practice and as a reproductive consultant to numerous Thoroughbred breeding farms before beginning a residency in equine reproduction at Auburn University in 2008. Dr. Dujovne completed her residency and Master of Science degree (studying the use of etonogestrel implantation for estrogen suppression in mares) in 2011, and remained at Auburn gaining experience as a clinical reproduction instructor. She is a diplomate of the American College of Theriogenologists. She joined the UC Davis School of Veterinary Medicine as an associate staff veterinarian and clinical professor in equine reproduction in 2012 and is currently an assistant professor in the Department of Population Health and Reproduction and chief of the equine reproduction service

Carrie Finno, DVM, Ph.D., DACVIM

Dr. Carrie Finno is an equine internist who received her DVM from the University of Minnesota. She then went on to complete a 3-year residency in large animal internal medicine at UC Davis, culminating in board certification in the American College of Veterinary Internal Medicine. Dr. Finno elected to pursue a career in equine genetic research, with a strong focus on neuromuscular disease, and obtained her Ph.D. in 2012 from UC Davis. Dr. Finno's research is focused on equine genetic diseases, including equine neuroaxonal dystrophy/equine degenerative myeloencephalopathy (NAD/EDM). In conjunction with the equine studies, she is researching the interaction of vitamin E and neural development, using a well-established mouse model. Dr. Finno was appointed as the director of the UC Davis Center for Equine Health in 2017 and is an associate professor in the Department of Population Health and Reproduction.

Larry Galuppo, DVM, DACVS

Dr. Larry Galuppo is a professor in the Department of Surgical and Radiological Sciences. He graduated from the UC Davis School of Veterinary Medicine in 1990 and completed an internship at Rood and Riddle Equine Hospital in 1991. He completed an equine surgery residency at UC Davis from 1991 to 1994, and he has been on the faculty at UC Davis since 1996. His area of clinical expertise is in equine orthopedic surgery, including tendon, ligament and joint disorders, with a special interest in traumatology and fracture repair. His research emphasis is on the biomechanics of fracture generation, implant design and fracture repair, with a recent focus in management of musculoskeletal injuries using regenerative medicine therapies in sport horses.

Robert Grahn, Ph.D.

Dr. Robert Grahn earned his doctorate from the University of Idaho, Moscow and spent 14 years investigating inherited genetic diseases in cats at UC Davis. He joined the UC Davis Veterinary Genetics Laboratory (VGL) in 2013, where he has assisted in the research and development of new test offerings for multiple species. He also works as an accredited forensic analyst within the VGL-Forensics section and mentors students in the Masters of Forensic science program at UC Davis. In 2019, he was appointed into the dual role of Associate Director of Service and Test Development. He and his team collaborate with scientists at UC Davis and around the globe to facilitate the advancement of important genetic discoveries. Dr. Grahn's own research efforts focus on the identification of novel genetic variants that can be used by animal owners, breeders, and veterinarians to make informed decisions regarding their animals.

Scott Katzman, DVM, DACVS

Dr. Scott Katzman received his DVM from the University of Minnesota, College of Veterinary Medicine. Following four years in private practice, he returned to academia to complete a three-year residency in equine surgery at the UC Davis School of Veterinary Medicine. He is a board certified diplomate of the American College of Veterinary Surgeons. Following completion of his surgical training, Dr. Katzman spent the following two years as the staff surgeon at an equine referral clinic in Minnesota, as well as working at a variety of equine referral practices across the country before joining the UC Davis faculty. Dr. Katzman has a special interest in musculoskeletal injury in racehorses and upper respiratory surgery. He is currently an assistant professor in the Department of Surgical and Radiological Sciences and chief of the equine surgery and lameness service.

Isabelle Kilcoyne, MVB, DACVS

Dr. Isabelle Kilcoyne earned her veterinary degree from the University of Dublin (Ireland) in 2008, after which she spent a year as an equine surgical intern at their University Veterinary Hospital. She then joined the UC Davis School of Veterinary Medicine, first as a team member with the Equine Field Service for two years, and then completed a three-year residency in equine surgery. Dr. Kilcoyne joined the Equine Surgical Emergency and Critical Care Service as an assistant clinical professor in 2016. Dr. Kilcoyne is a board certified diplomate in the American College of Veterinary Surgeons. Her main clinical and research interests are in emergency surgery and medicine, particularly gastrointestinal surgery.

Heather Knych, DVM, Ph.D., DACVCP

Dr. Heather Knych is a professor of Clinical Veterinary Pharmacology. She earned her veterinary degree at the UC Davis School of Veterinary Medicine , followed by her Ph.D. in pharmacology. She is a diplomate of the American College of Veterinary Clinical Pharmacology. Dr. Knych's research focuses on equine drug metabolism and pharmacokinetic/pharmacodynamics (PK/PD) relationships of drugs in performance horses. Additionally, Dr. Knych provides guidance to researchers at UC Davis and other universities as well as to drug companies on PK/PD study design. She assists with drug concentration determination and pharmacokinetic analysis in various biological matrices.

Mary Lassaline, DVM, Ph.D., MA, DACVO

Dr. Mary Lassaline received her veterinary training at Michigan State University. She completed an internship in equine medicine and surgery at Rood and Riddle Equine Hospital in Lexington, Kentucky and a residency in comparative veterinary ophthalmology at the University of Florida. She worked in private ophthalmology practice for two years in Fairfield County, Connecticut before joining the faculty at the University of Pennsylvania's New Bolton Center, where she started a full time equine ophthalmology service. She joined the ophthalmology team at UC Davis to support the expansion of the large animal ophthalmology service. Dr. Lassaline is a diplomate of the American College of Veterinary Ophthalmologists. She is Chair of the American Board of Veterinary Ophthalmologists Credentials Committee and is engaged in clinical research on equine corneal disease.

John Madigan, DVM, MS, DACVIM

Dr. John Madigan is a distinguished professor of medicine and epidemiology at the UC Davis School of Veterinary Medicine and a diplomate of the American College of Veterinary Internal Medicine and the American College of Animal Welfare. He is a clinician in equine medicine at the Veterinary Medical Teaching Hospital where he started the UC Davis Veterinary Medical Teaching Hospital Equine Neonatal Critical Care Unit in 1987. He leads the Comparative Neurology Research Group at UC Davis investigating neurological conditions of horses and humans. Dr. Madigan is a recipient of a Bill and Melinda Gates Foundation grant with Stanford Medical School and UC Davis School of Medicine investigating transition of consciousness at birth in infants based on recent discoveries in neonatal foals.

K. Gary Magdesian, DVM, DACVIM, DACVECC, DACVCP

Dr. Gary Magdesian received his DVM from the UC Davis School of Veterinary Medicine and completed an internship in large animal medicine and surgery at the College of Veterinary Medicine at Texas A&M University. He then completed residencies in equine internal medicine, equine emergency medicine/critical care and clinical pharmacology at the School Veterinary Medicine, UC Davis. Dr. Magdesian is board certified in internal medicine, emergency/critical care and pharmacology. Currently, Dr. Magdesian is a professor in the Department of Medicine and Epidemiology and holds the Roberta and Carla Henry Endowed Chair in Emergency Medicine and Critical Care.

Beatriz Martínez López, DVM, MPVM, Ph.D.

Dr. Beatriz Martínez López received her veterinary degree from Complutense University, Madrid, Spain, in 2004 and her MPVM from the University of California, Davis in 2007. She earned a doctorate degree from Complutense University, Madrid, Spain in 2009. Dr. Martínez López is an associate professor in the Department of Medicine and Epidemiology and holds a faculty appointment with the Agricultural Experiment Station. She is also the Director of the UC Davis Center for Animal Disease Modeling and Surveillance (CADMS). Her research is focused on the development and application of epidemiological tools for supporting more cost-effective and risk-based surveillance and control strategies. She has primarily been working on epidemiological modeling and risk assessment for the evaluation of the potential introduction and/or spread of diseases affecting domestic and/or wild animal populations, many of which are considered to be emerging or re-emerging due to globalization, changes in climate and land use.

Stuart Meyers, DVM, Ph.D., DACT

Dr. Stuart Meyers, a professor in the Department of Anatomy, Physiology, and Cell Biology, earned his veterinary degree from the University of Michigan in 1985 and his Ph.D. in comparative pathology from UC Davis in 1995. He is a diplomate of the American College of Theriogenologists. Dr. Meyers' research focuses on membrane and cytosolic events associated with sperm cell function and developing methods by which sperm preservation and fertility can be advanced. The laboratory is examining the role of membrane lipid domains and their associated proteins relative to sperm capacitation, osmotic and oxidative stress, and cryopreservation. Studies are aimed at optimization of male genome preservation and understanding of mechanisms of male subfertility.

Michael Mienaltowski, DVM, Ph.D.

Dr. Michael Mienaltowski earned his DVM degree from Michigan State University in 2004, followed by his Ph.D. at the University of Kentucky in 2008. He continued with post-doctoral training at the University of South Florida from 2008 through 2014, with a focus on molecular pharmacology and physiology, orthopedics and sports medicine. He joined the faculty of the UC Davis College of Agricultural and Environmental Sciences in 2014 and is currently an assistant professor in the Department of Animal Science.

Jessica Morgan, DVM, Ph.D., DACVSMR

Dr. Jessica Morgan joined the Equine Field Service as an assistant professor in 2019 and currently serves as chief of service. Dr. Morgan received her PhD (2012) and DVM (2013) from UC Davis. She completed an internship at Peninsula Equine Medical Center in Menlo Park, California, and a three-year residency in equine sports medicine and rehabilitation at the University of Pennsylvania School of Veterinary Medicine, New Bolton Center. Dr. Morgan remained at New Bolton Center as a lecturer in equine exercise physiology and then as a lecturer in large animal ultrasound and cardiology. She is a diplomate of the American College of Veterinary Sports Medicine and Rehabilitation. Dr. Morgan's research interests and expertise are in basic and applied science related to equine performance, musculoskeletal disease, and lameness diagnosis, including early detection and treatment of performance limiting conditions of horses and characterization of the roles that matricellular proteins play in tissue degeneration and disease prediction.

Nicola Pusterla, DVM, Ph.D., DACVIM

Dr. Nicola Pusterla graduated from the School of Veterinary Medicine at the University of Zurich, Switzerland in 1991. Dr. Pusterla worked in the private and academic sector with a focus in large animal internal medicine and earned his Ph.D. from the University of Zurich with an emphasis on vectorborne diseases. He joined UC Davis in 1998 where he currently has an appointment as a professor in Equine Internal Medicine in the Department of Medicine and Epidemiology. Dr. Pusterla is a diplomate of the American College of Veterinary Internal Medicine with an equine emphasis, and he has ongoing interest in all aspects of equine internal medicine and dentistry. Dr. Pusterla's research focuses on selected aspects of equine infectious diseases with an emphasis on epidemiology, clinical disease understanding, diagnostics, prevention, and treatment.

Jeroen Saeij, Ph.D.

Dr. Jeroen Saeij earned his master's and doctorate degrees from Wageningen University, Wageningen, The Netherlands. He completed a postdoctoral fellowship at Stanford University before becoming a professor at the Massachusetts Institute of Technology. He came to UC Davis in 2015 and is now an associate professor in the Department of Pathology, Microbiology & Immunology. Dr. Saeij's research focus is on the molecular basis of pathogenesis of Apicomplexan parasites in humans and livestock. He is interested in how the interactions between parasite and host can lead to disease with a special interest in the genetic basis for individual host differences in resistance and parasite differences in virulence.

Joao Soares, MV, MSc, DSc, DACVAA

Dr. Joao Soares joined the Anesthesia/Critical Patient Care Service as an assistant professor in 2018. He received his DVM in 1999 and his MSc in 2002 from the Fluminense Federal University, Brazil and his doctorate in 2012 from the Rio de Janeiro Federal University. He completed his residency in 2012 in veterinary anesthesiology at UC Davis and continued as a staff veterinarian from 2012-2014. Dr. Soares was an assistant professor of anesthesiology from 2014-2018 at the Virginia-Maryland Regional College of Veterinary Medicine. His research focuses on monitoring respiratory function during anesthesia, including linear and nonlinear models of respiratory mechanics, volumetric capnography, and electrical impedance tomography, and on protective ventilation during anesthesia including monitoring ventilator settings and effects on outcome.

Sharon Spier, DVM, Ph.D., DACVIM

Dr. Sharon Spier, a professor emeritus in the UC Davis School of Veterinary Medicine, earned her veterinary degree from Texas A&M University in 1983 and her Ph.D. in Comparative Pathology from the University of California, Davis in 1989. She is diplomate of the American College of Veterinary Internal Medicine. Dr. Spier's research focus is on Corynebacterium pseudotuberculosis infections in horses and she pioneered the understanding of hyperkalemic periodic paralysis in horses.

Mathieu Spriet, DVM, MS, DACVR, DECVDI, DACVR-EDI

Dr. Mathieu Spriet graduated from the National Veterinary School of Lyon, France in 2002. He completed an equine internship and master's degree at the University of Montreal (Canada). He completed his residency in diagnostic imaging at the University of Pennsylvania in 2007. Dr. Spriet is currently an associate professor of Clinical Diagnostic Imaging at the UC Davis School of Veterinary Medicine. He is a diplomate of both the American College of Veterinary Radiology and the European College of Veterinary Diagnostic Imaging. He recently became a diplomate of the newly recognized ACVR sub-specialty for Equine Diagnostic Imaging (2019). His main research interest is on musculoskeletal imaging. He has pioneered the use of Positron Emission Tomography in horses, leading to the development of an equine specific PET scanner.

Joshua Stern, DVM, Ph.D., DACVIM

Dr. Joshua Stern is an associate professor and residency program director for cardiology at the UC Davis School of Veterinary Medicine. He operates one of only a few translational cardiac genetic laboratories in the world. Together with a team of graduate students, he studies the intersection of genetics and cardiac pharmacology and focuses on mutation discovery and pathogenesis for inherited heart disease. Dr. Stern serves as the chair of the board examination committee for the American College of Veterinary Internal Medicine subspecialty of cardiology. Through his work with the cardiology service at UC Davis, Dr. Stern leads innovative interventional procedures, identifies novel diagnostic approaches and carries out clinical studies for novel therapeutics.

Susan Stover, DVM, Ph.D.

Dr. Susan Stover is a professor in the Department of Surgical and Radiological Sciences and director of the J.D. Wheat Veterinary Orthopedic Research Laboratory at the UC Davis School of Veterinary Medicine. She received her veterinary degree from Washington State University, and subsequently completed an equine surgery internship and residency at UC Davis. She was in equine practice in Washington State before returning to UC Davis to teach clinical equine lameness and surgery to veterinary students and residents. She became board certified by the American College of Veterinary Surgeons while pursuing a Ph.D. program focused on equine orthopedic research (dorsal metacarpal disease ('bucked shins') in Thoroughbred racehorses). Dr. Stover now devotes her time to equine orthopedic research and teaches musculoskeletal anatomy, biomechanics, and pathology to veterinary students. She was honored with the American Veterinary Medical Association Lifetime Excellence in Research Award in 2016.

Fern Tablin, VMD, Ph.D.

Dr. Fern Tablin, professor emeritus, earned her VMD in 1980 and her Ph.D. in 1984 from the University of Pennsylvania. She joined the UC Davis School of Veterinary Medicine faculty in 1985, with a research focus on platelet physiology and the role of platelets in health and disease, with a concentration on the contribution of platelets to the systemic, cardiac and pulmonary proinflammatory responses to air pollution. Serving as co-director for the Center for Biostabilization, Dr. Tablin developed novel methods for storage of blood cells and nucleated cells in the dry state. She was inducted into the American Association for the Advancement of Science in 2002, and was awarded the Pfizer Distinguished Teacher Award in 2012.

Alain Théon, DVM, MS, Ph.D., DACVR

Dr. Alain Théon received his DVM from Ecole Nationale Vétérinaire d'Alfort, (Maisons-Alfort France). He completed a 3-year research doctorate program in Radiation Biology at University Paris-Est (Creteil, France) concurrently with a 2-year internship in Radiation Oncology at Tenon Hospital (University Medical Center, Paris, France). He moved to the United States to pursue a training program in veterinary radiation oncology and completed a 2-year limited-status residency in Therapeutic Radiology at the UC Davis School of Veterinary Medicine. Dr. Théon completed a M.S. degree in Comparative Pathology at UC Davis funded by the Center for Companion Animal Health and joined the UC Davis School of Veterinary Medicine faculty in 1990. Since then he has dedicated his career to teaching and research that benefits dogs, cats and horses with cancer.

Joie Watson, DVM, Ph.D., DACVIM

Dr. Joie Watson received her veterinary degree in 1986 and her Ph.D. in 1994 from the UC Davis School of Veterinary Medicine and joined the faculty in 1996 as a professor of equine medicine. She is a diplomate of the American College of Veterinary Internal Medicine. Her research focus is in immunology and infectious diseases in horses. In 2014, she was honored with the UC Davis School of Veterinary Medicine's Faculty Distinguished Teaching Award.

David Wilson, BVMS, Hon DACVIM, MRCVS

Dr. David Wilson, professor emeritus, graduated from Glasgow University as a member of the Royal College of Veterinary Surgeons in 1975. He obtained a Master of Science Degree in Medicine and Immunology from Iowa State University, where he was an instructor and professor from 1975 - 1981. In 1981, he began his 36-year tenure at the University of California, Davis, where he held various positions, including: director of the Center for Equine Health, director of the William R. Pritchard Veterinary Medical Teaching Hospital (VMTH), and associate dean of Clinical Programs. In 2019, he received the American Association of Equine Practitioners Distinguished Educator – Academic Award. He received the 1989 Norden Distinguished Teacher Award and the 2013 Honorary Diplomate Award of the American College of Veterinary Internal Medicine for his contributions to the discipline of Large Animal Internal Medicine. Throughout his career, Dr. Wilson advanced the area of equine internal medicine and infectious diseases.

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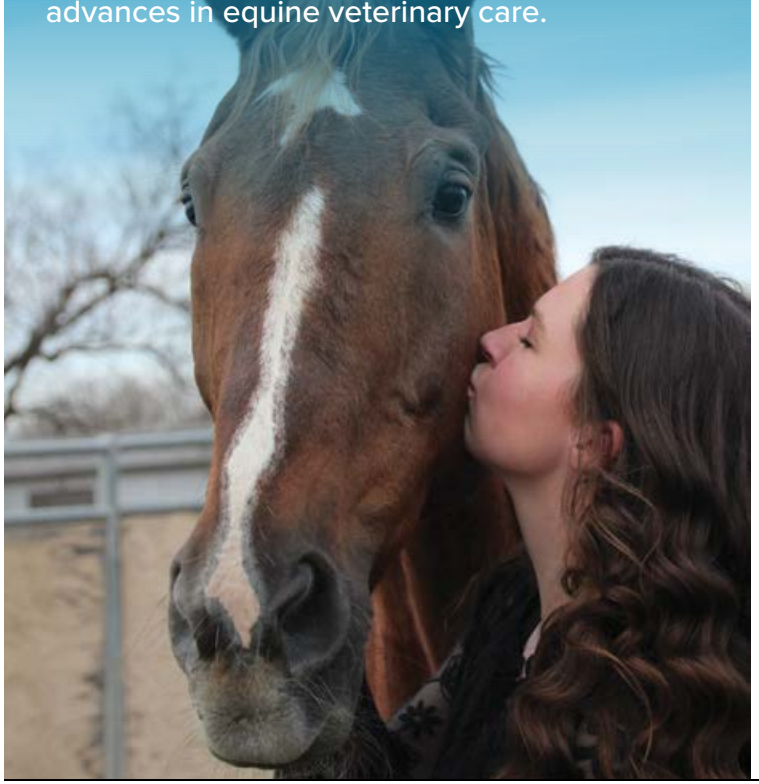
The Equine Tribute and Memorial Fund provides a meaningful way to pay tribute to special horses and the people that love them. Participating veterinarians, horse owners, friends, and family can honor horses that have passed, support those that have lost beloved horses, celebrate the birth of new foals, commemorate performance accomplishments, and pay tribute to individuals and organizations. Donations to this fund support cutting-edge equine research and valuable advances in equine veterinary care.



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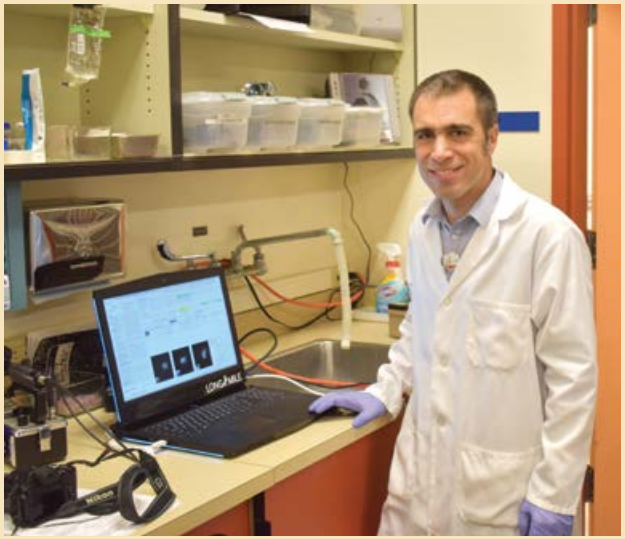
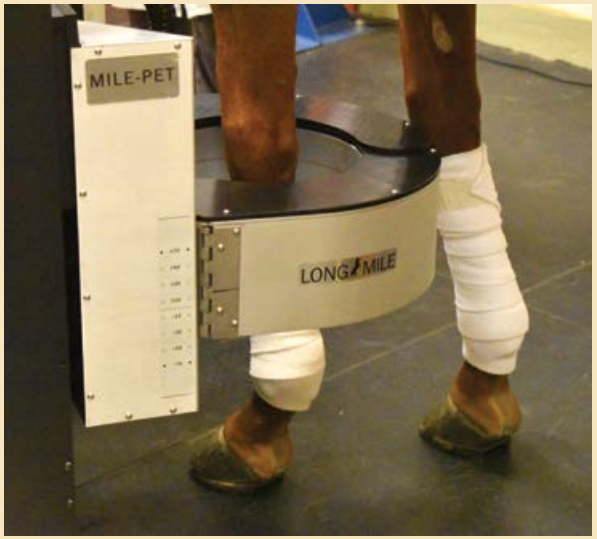
EQUINE PET: A GAME CHANGER

In 2015, researchers at the UC Davis School of Veterinary Medicine performed the first equine PET scan. Less than five years later, this cutting-edge technology is being used on a daily basis at California's Santa Anita racetrack to investigate bone changes in racehorses in an effort to prevent injuries. Support from the Center for Equine Health played a critical role in getting it all started.

"It's really exciting to see how quickly this research has been put into clinical use," said Dr. Mathieu Spriet, associate professor of diagnostic imaging at UC Davis and leader of the equine PET research efforts. "It started as a very broad concept to see what we could do, but now we have defined several pertinent opportunities for clinical use."



Dr. Mathieu Spriet



LONGMILE MILEPET standing equine PET scanner

Positron emission tomography (PET) is an advanced imaging technology similar to a "bone scan" (scintigraphy). However, PET provides three-dimensional (3D) information whereas traditional bone scans project anatomy in two dimensions. To capture data in 3D, the PET scanner uses a ring of detectors that fully circle the horse's leg. Initially, this meant that horses had to be lying down on a table under general anesthesia to undergo a scan.

To overcome barriers associated with anesthesia, Spriet collaborated with LONGMILE Veterinary Imaging to design the MILEPET (Molecular Imaging of Limbs in Equids), which features an openable ring of detectors to image standing horses. The instrument was installed at Santa Anita racetrack in December of 2019.

"It's above and beyond all expectations," said Dr. Joseph Dowd, president of the Southern California Equine Foundation, which owns and operates the PET unit. "It showed up on time, on budget, and did exactly what they said it would do. They began scanning horses the day after it was assembled on site."

In the first nine months after installation, more than 150 studies were performed on close to 100 horses sent in by 15 different veterinarians for over 30 different trainers. "The trainers call it a game changer," said Dowd. "It really allows us to dial in and see exactly where these areas of pathology are."

To date, the standing PET has been used to image horses' legs from the foot to the knee, with particular emphasis on the fetlock, where the proximal sesamoid bones are a common fracture site in racehorses (see Dr. Stover's research summaries in this report - grant # 17-04S and 18-04).

"PET has been excellent at identifying abnormalities in the sesamoid bones that bone scan and radiographs missed," said Spriet. "Also, PET can distinguish different categories of injuries in the sesamoid bones, which lead to differences in case management."

PET has high sensitivity so it can detect early and subtle changes in bone and/or soft tissue, depending on the tracer used. It is a "functional" technique, meaning that it can highlight active injuries, whereas other imaging modalities may not be able to distinguish active from inactive injuries. PET can be combined with MRI or CT information to provide anatomical detail.

"From my point of view, its greatest asset is as a screening tool," said Dowd. "I think we're very close to a high throughput screening technique that will allow us to identify horses at risk and move them into a rehabilitation program or send them for further diagnostics."

Studies funded by the Grayson Jockey Club and Dolly Green Foundation are using the standing PET to follow racehorses over time. These studies involve sequential scans on the same horses over a few months as well as following horses diagnosed with an injury by PET back into training after a period of rehabilitation.

"Things can change quickly on PET," said Spriet. "We have seen that, with rest, a fair number of injuries resolved in six to 12 weeks, while others persisted. This shows that PET is a great tool to adapt treatment and rehabilitation plans."

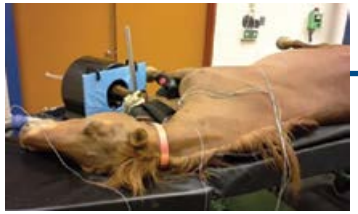
"This has been an overwhelming success, above and beyond what we ever expected," said Dowd. "I think it will sweep the horse racing world, for sure. Everyone will have these at any high level racetrack."

In addition to racehorse use, research at UC Davis has been performed in sport horses, with studies on joint disease, laminitis and deep digital flexor tendon lesions. As more data is collected and additional instruments are installed (the University of Pennsylvania recently acquired one), Spriet is confident that applications for the technology will expand.

"What began merely as an academic curiosity has progressed quickly," reflected Spriet. "The reasons for this success are that we had the appropriate support and environment at UC Davis, thanks to CEH."

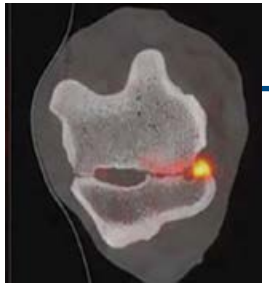
EQUINE PET TIMELINE

2015 — First equine PET scan performed at UC Davis

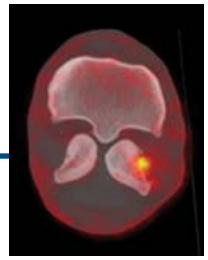


2016 — UC Davis first veterinary hospital in the world to install an equine PET scanner for clinical cases

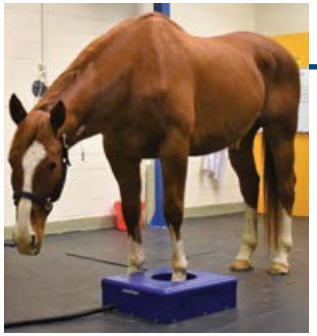
2017 — UC Davis begins clinical trials using the equine PET scanner



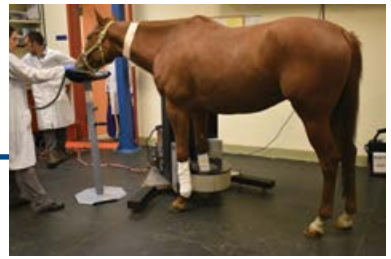
2018 — UC Davis researchers announce completion of more than 85 equine PET studies and validation of dual-tracer scanning protocol



2019 — First standing equine PET scan performed at UC Davis using the scanner prototype

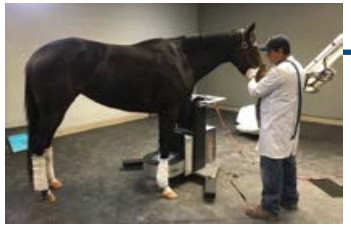


2019 — First standing equine PET scan with MILE-PET scanner

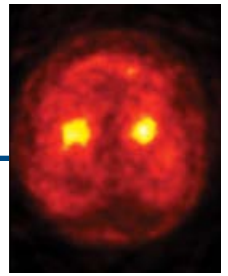


2019 — Stronach Group announces investment to support the purchase of the MILE-PET scanner

2019 — Standing MILE-PET scanner installed at Santa Anita Racetrack



2020 — Longitudinal studies in racehorses at Santa Anita racetrack



We value your partnership in our mission to improve the health and well-being of horses.

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